VIRULENCE, PATHOGENICITY, AND HOST SPECIFICITY OF *SALMONELLA ENTERICA SUBSPECIES ENTERICA* SEROVARS

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VIRULENCE, PATHOGENICITY, AND HOST SPECIFICITY OF *SALMONELLA ENTERICA SUBSPECIES ENTERICA* SEROVARS

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Salmonella is a gram-negative zoonotic foodborne pathogen and the etiologic agent of salmonellosis. Estimates indicate that nontyphoidal *Salmonella* is the leading bacterial cause of domestically acquired foodborne illnesses, hospitalizations, and deaths in the U.S. each year. This pathogen is known to have an extensive repertoire of virulence genes, and the disease severity could vary depending on the host and the bacterial strain. The studies presented herein were aimed at characterizing *Salmonella* serovars from different sources using phenotypic, genotypic and next generation sequencing approaches. Specifically, this work focuses on: (i) the diversity of *Salmonella* subtypes from subclinical dairy cattle and farm environments, (ii) the genomic features of *Salmonella* Cerro, the most commonly isolated serovar from the farms studied here, and (iii) the typhoid toxin (CdtB), a virulence factor encoded in a genomic islet in nontyphoidal *Salmonella* serovars, and its effects on the cell cycle of human epithelial cells (Henle-407). Our data suggest that subclinical livestock and farm environments may represent important reservoirs of *Salmonella* serovars and subtypes associated with human infections, including MDR *Salmonella.* Interestingly, serovars that are rare among human clinical cases were found in subclinically infected cattle and farm environments (e.g., *Salmonella* Cerro). Our comparative and population genomic analyses of 27 *Salmonella* Cerro genomes revealed that the increase in prevalence of this serovar in dairy cattle is caused by a highly clonal subpopulation which represent

sequence type ST367. Furthermore, genomic deletions in *Salmonella* Cerro ST367 may indicate adaptation to specific ecological niches and possibly reduced virulence in some hosts. Finally, we showed that the nontyphoidal CdtB is closely related to the *Salmonella* Typhi CdtB and also has a possible role in host-pathogen interaction since Henle-407 cells infected with wild type strains displayed arrest in G_2/M, while cells infected with the isogenic mutants for *cdtB* did not. Altogether, these studies contribute to our current understanding of: (i) *Salmonella* serovars and subtypes associated with subclinical hosts and farm environments (ii) genomic factors that may contribute to nontyphoidal *Salmonella* adaptation to certain hosts, and (iii) virulence factors, specifically the typhoid toxin CdtB, and its role in the pathogenesis of nontyphoidal *Salmonella* serovars.

BIOGRAPHICAL SKETCH

Lorraine holds a bachelor of science in Microbiology from the University of Puerto Rico. She was an undergraduate research assistant in the Geomicrobiology Laboratory and the Cabo Rojo Salterns NSF Microbial Observatory directed by Dr. Lilliam Casillas-Martinez. After taking a Food Microbiology course, she developed an interest in food safety, and soon after was selected to participate in the Cornell Summer Scholars Program in 2007. A year after, she was admitted to the graduate program in Food Science at Cornell University. Since then she has worked on her research under Dr. Wiedmann in the Laboratory of Food Microbiology and Pathogenesis of Infectious Diseases. Her Ph.D. thesis research focused on the virulence, pathogenicity and host specificity of *Salmonella enterica*. Through collaboration with research groups inside and outside Cornell, she is a co-author and first author on 12 manuscripts, 10 of which have been published in peer-reviewed journals and two additional ones are either accepted for publication or submission-ready. She is currently a Postdoctoral Research Associate in Dr. Cumming's laboratory at Texas A&M University. Her career goal is work as a professor in Puerto Rico and to serve as an inspiration for minority students to encourage them to pursue careers in Food Safety.

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CHAPTER 1

INTRODUCTION

Salmonella is a zoonotic foodborne pathogen and the etiologic agent of salmonellosis. Salmonellosis is one of the leading foodborne illnesses in the United States. It has been estimated that approximately 1.0 million nontyphoidal *Salmonella* illnesses, 20,000 hospitalizations and 370 deaths occur through foodborne transmission in the U.S. each year [1]. Furthermore, 93.4 million gastroenteritis cases, and 155,000 deaths are estimated to be caused by salmonellosis each year worldwide [2].

Salmonella comprises of two species, *Salmonella bongori* and *Salmonella enterica*, and more than 2,600 recognized serovars [3]. However, most human salmonellosis cases are caused by relatively few serovars within *S. enterica*, and disease severity or outcome could vary by serovar [4].

Salmonella serovars have different hosts and reservoirs ranging from cold-blooded (e.g., reptiles) to warm-blooded animals (e.g., mammals) [5]. Furthermore, *Salmonella* can survive in farms and other environments for prolonged periods of time [6, 7]. *Salmonella* is transmitted to animals and humans through the fecal-oral route. Humans can become infected by foodborne transmission or after direct or indirect contact with infected animals [5],. Disease manifestations in people include diarrhea, fever, and abdominal cramps, appearing 12 to 72 hours after ingestion. *Salmonella* infection in humans usually results in self-limiting gastroenteritis although in severe cases it can spread systemically. Animals can become infected after ingestion of feed and water contaminated with *Salmonella*. In livestock, clinical signs typically appear 6 to 24 hours after exposure and include profuse diarrhea, fever, dehydration, inappetence, foul-smelling feces, and mucus or blood in feces [8]. *Salmonella* can also be carried subclinically by both humans and animals [9–11].

While serovars mostly associated with human clinical cases have been studied and characterized extensively (e.g., Typhi, Typhimurium, and Enteritidis), more emphasis on non-

human associated serovars is needed to avoid bias in the understanding of the biology, ecology, and genetics of *Salmonella* spp.

CHARACTERIZATION OF SALMONELLA SPP.

Subtyping Approaches

Different subtyping methods have been used to characterize *Salmonella* beyond the subspecies and serovar level. Multilocus Sequence Typing (MLST) is a very useful tool for characterizing *Salmonella* isolates, and has been proposed as an option for replacing serotyping [12]. In general, this method consists on using the internal fragments of seven housekeeping genes to classify strains into Sequence Types (ST) and those STs are used to study the genetic relationship between the strains. Variations of this method have been described in the past [13, 14]. One of the advantages of MLST is that the data on many different microorganisms is publicly available (http://pubmlst.org/databases.shtml, and http://www.genomicepidemiology.org/). In addition, Pulsed-Field gel electrophoresis (PFGE) is the "gold-standard" method currently used by Pulsenet, Centers for Disease Control and Prevention (CDC) to subtype bacterial pathogens during outbreak investigations. PFGE typing consists on separating large fragments of bacterial DNA, obtained from digestion with restriction endonucleases, on an agarose gel subjected to an electric field directed in different directions [15]. Some of the limitations of this method are that it is time consuming and requires well-trained staff to obtain quality data. Multilocus variable-number of tandem-repeat analysis (MLVA) consists of amplifying fragments of DNA that contain tandem repeats, quantifying the number of repeats in each locus, and finally assigning a code. The string of code is called an MLVA type. So far, specific MLVA analyses have been developed for the study of several pathogens including *Salmonella enterica* subsp. *enterica* serovars (i.e., Typhimurium, Enteritidis, Typhi, Infantis) [16–19]. This method is also used by Pulsenet (CDC) to investigate outbreaks linked to *S.* Typhimurium, and *S.* Enteritidis, and it has the potential to be a great subtyping method, but more standardized protocols have to be developed.

Sequencing Approaches

Recent studies have characterized a larger and more diverse group of *Salmonella* serovars using comparative and population genomics as a tool [20–24]. Some of those studies have demonstrated that *Salmonella* is divided into different subpopulations [20, 21]. Several studies have shown that next generation sequencing approaches have proven to be successful for the study of foodborne outbreaks caused by different pathogens (i.e., *E. coli*, *Vibrio*) [25, 26], including *Salmonella* [24, 27, 28].

Studies which have used a combination of *Salmonella* genetic modifications (e.g., null mutations, transposon mutagenesis) and phenotypic assays of the mutants (e.g., *in vitro* and *in vivo* infection experiments) have been crucial for the understanding of *Salmonella* pathogenesis, virulence, and host specificity. Most of the virulence genes and *Salmonella* pathogenicity islands (SPIs) have been discovered and characterized using the aforementioned approaches [29–33].

Our study

To gain a broader knowledge of *Salmonella* diversity, virulence, and host specificity, several approaches were used in the studies conducted as part of this dissertation. A total of 1,349 *Salmonella* isolates were obtained from subclinically infected dairy cattle and dairy farm environments in 46 NY state farms. All of these isolates were characterized by serotyping and antimicrobial susceptibility assays, and a subset (*n = 381*) were PFGE typed [34] The most common serovar isolated from the farms in our study was *Salmonella* Cerro, which has been described as an emerging pathogen among dairy cattle in the Northeastern US in the last few years [35–37], and it is one of the most commonly isolated serovars from clinically healthy dairy cattle, not only in the Northeastern US but throughout the country [10, 38]. *Salmonella* Cerro isolates represented a highly clonal population based on PFGE analysis, with 90.5% of our isolates sharing the same PFGE type, which is consistent with previous studies [35–37, 39]. *S.* Cerro is rarely associated with human disease, with only 1 known outbreak reported by the CDC in 1985 [40]. This serovar was reported to be frequently isolated from subclinical human carriers in Southern Italy [41], as well as from different animal species (e.g., reptile, insects, wild animals) [41, 42]. In order to investigate the distinctive epidemiological features (i.e., frequent

isolation from dairy cattle but rare association with human disease) 27 *S*. Cerro genomes were sequenced using the SOLiD™ platform (Life Technologies).

In addition to the *S*. Cerro sequencing project, a *Salmonella* sequencing project led by our research group [20] has shown that *Salmonella* could be classified into two distinct subpopulations, or clades designated as clade A and clade B. These clades possess clade-specific genes that have been described as being responsible for differences in adhesion, colonization, and metabolic capabilities between *S. enterica* subsp. *enterica* clades [20]. The pathogenicity islet encoding the typhoid toxin, known as the cytolethal distending toxin B islet (CdtB-islet) was found in nontyphoidal *Salmonella* serovars that mostly belong to clade B. This pathogenicity islet comprises five genes (i.e., *pltA*, *pltB*, *ttsA*, *sty1887*, in addition to *cdtB*). CdtB is a recently recognized virulence factor that is found in *Salmonella enterica* serovar Typhi [43, 44] as well as a number of other Gram-negative bacterial pathogens [45]. Host cells intoxicated with CdtB undergo irreversible cell cycle arrest in response to DNA damage, which can lead to cell death by apoptosis [45]. We hypothesized that CdtB, in clade B nontyphoidal *Salmonella* strains, is functional and causes G_2/M cell cycle arrest, similar to the CdtB effect reported for *Salmonella* Typhi [44, 46]. To address our hypothesis we used a comparative genomics approach to characterize sequence conservation of *plt*A, *plt*B, and *cdt*B among *Salmonella* Typhi and nontyphoidal *Salmonella* serovars. In order to assess the function of CdtB in three nontyphoidal *Salmonella* strains, we created three CdtB isogenic null mutants, and infected Henle-407 cells with all 6 strains. Then, host cell cycle analysis by Fluorescence-Activated Cell Sorting (FACS) was performed.

More studies are needed to elucidate the underlying mechanisms and factors that make *Salmonella* serovars so successful in colonizing and infecting a number of hosts, and surviving in different environments. Although many virulence and genomic factors have been described, there is still a great amount of work that needs to be done in order to address the lack of knowledge about most of the *Salmonella* serovars. The studies described as part of this dissertation add to the growing body of knowledge about *Salmonella* serovars and contribute to

the understanding of the evolution, virulence, and host specificity of *Salmonella*. Findings generated by future studies, with more comprehensive data sets, will allow for better tracking of outbreaks and for the development of better diagnostic tools in order to mitigate and control this pathogen.

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CHAPTER 2

SUBTYPE ANALYSIS OF *SALMONELLA* ISOLATED FROM SUBCLINICALLY INFECTED DAIRY CATTLE AND DAIRY FARM ENVIRONMENTS REVEALS THE PRESENCE OF BOTH HUMAN- AND BOVINE-ASSOCIATED SUBTYPES*

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ABSTRACT

While it is well established that clinically ill livestock represent a reservoir of *Salmonella*, the importance of subclinical shedders as sources of human salmonellosis is less well defined. The aims of this study were to assess the subtype diversity of *Salmonella* in healthy dairy cattle and associated farm environments and to compare the subtypes isolated from these sources with the *Salmonella* subtypes associated with clinical human cases in the same geographic area. A total of 1,349 *Salmonella* isolates from subclinical dairy cattle and farm environments (46 farms) were initially characterized by traditional or molecular serotyping and tested for antimicrobial susceptibility. A set of 381 representative isolates was selected for further characterization by pulsed-field gel electrophoresis (PFGE); these isolates represented unique combinations of sampling date, serovar, antimicrobial resistance pattern, farm of origin, and source, to avoid overrepresentation of subtypes that were re-isolated from a given source. These 381 isolates represented 26 *Salmonella* serovars; the most common serovars were Cerro [(38.8%, 148/381) isolated from 21 farms], Kentucky [16.3%; 10 farms], Typhimurium [9.4%; 7 farms], Newport [7.6%; 8 farms], and Anatum [6.3%; 6 farms]. Among the 381 isolates, 90 (23.6%) were resistant to between 1 and 11 antimicrobial agents, representing 50 different antimicrobial resistance patterns. Overall, 61 *Xba*I-PFGE types were detected among these 381 isolates, indicating considerable *Salmonella* diversity on dairy farms without evidence of clinical salmonellosis. Fourteen PFGE types, representing 12 serovars, exactly matched PFGE types from human isolates, suggesting that subclinically infected dairy cattle could be sources of human disease-associated *Salmonella*.

INTRODUCTION

Salmonella is a zoonotic foodborne pathogen and the etiologic agent of salmonellosis. Salmonellosis is a major health concern as it is one of the leading causes of foodborne illness in the United States. It has been estimated that approximately 1.0 million nontyphoidal *Salmonella* illnesses, 20,000 hospitalizations and 370 deaths occur through foodborne transmission in the

U.S. each year [1]. *Salmonella* is comprised of two species, *Salmonella bongori* and *Salmonella enterica*, and more than 2,600 recognized serovars [2]. However, most human salmonellosis cases are caused by relatively few serovars within *S. enterica* [3]. *Salmonella* serovars have different hosts and reservoirs ranging from cold-blooded (e.g., reptiles) to warm-blooded animals (e.g., mammals) [4]. Furthermore, *Salmonella* can survive in farm and other environments for prolonged periods of time [5, 6]. Some previous studies have established that certain serovars may be overrepresented among specific hosts and/or associated with specific hosts; for example, while serovar Dublin has been isolated from both bovine and human hosts, it is most commonly isolated from cattle and rarely found in other non-primate hosts and is thus typically considered "bovine associated".

Salmonella is transmitted to animals and humans through the fecal-oral route. Animals can become infected after ingestion of feed and water contaminated with *Salmonella.* Similarly, humans can become infected by foodborne transmission or after direct or indirect contact with infected animals [4]. In livestock, clinical signs typically appear 6 to 24 hours after exposure and include profuse diarrhea, fever, dehydration, inappetence, foul-smelling feces, and mucus or blood in feces [7]. Disease manifestations in people include diarrhea, fever, abdominal cramps and septicemia in severe cases, appearing 12 to 72 hours after ingestion. *Salmonella* can also be carried subclinically by both humans and animals [8–10]. The purpose of this study was to investigate the phenotypic and genotypic diversity and distribution of *Salmonella* serovars isolated from subclinically infected cattle and associated farm environments within New York dairy farms.

MATERIALS AND METHODS

Study design

Bovine fecal and environmental samples were collected from dairy farms in New York between October 2007 and August 2009 as described by Cummings et al. [11]; 44 of the farms detailed in that study [11] as well as 2 additional New York dairy herds included here yielded

Salmonella-positive samples, for a total of 46 farms with *Salmonella*-positive samples. Briefly, *Salmonella* surveillance included both environmental screening and disease monitoring within each herd for a period of at least 12 months. A positive *Salmonella* culture result arising from either surveillance method would trigger subsequent visits for cattle sampling, and 50–70 fecal samples were collected from apparently healthy cattle at each visit depending on herd size. Overall, 8,948 samples (1,420 environmental and 7,528 fecal) were collected from the 46 dairy herds.

All *Salmonella* isolates obtained from the 46 farms were used here for phenotypic characterization (i.e., serotyping and antimicrobial susceptibility testing, as detailed below), totaling 1,349 isolates obtained from environmental (n = 402) and fecal samples (n = 947) (see Appendix Table 1 for a list of all isolates). Isolates were obtained over multiple sample collections at the same farm, with a median number of 4 sample collection dates with *Salmonella* positive samples per farm (range: 1–18); the wide range reflects that some farms might have only had a single sample collection that yielded *Salmonella*, while others might have had a large number of sample collections that yielded *Salmonella* (for details see [11]).

Salmonella isolation from environmental and bovine fecal samples

Salmonella isolation procedures have previously been reported [6, 7, 11]. Briefly, environmental samples were taken from four different locations on each farm (i.e., calf housing, cow housing, sick pen, and manure storage area), using sterile 4x4 gauze swabs saturated in double-strength skim milk. Gauze swabs were placed into a sterile flip-top container, and samples were stored at 4°C and brought to the laboratory for *Salmonella* isolation. Fecal samples from cattle without clinical signs of salmonellosis (i.e., animals that do not show diarrhea, fever, etc.) were collected via rectal retrieval using a new sleeve for each sample. Ten grams of fecal matter were placed into a Para-Pak bottle, sealed, and sent to the laboratory for culture and further analyses. Cultural testing of these environmental and fecal samples yielded 1,349 *Salmonella* isolates that were included in the study reported here.

Serotyping

Traditional serotyping was performed by agglutination [12] at the National Veterinary Services Laboratories (NVSL), a division of the United State Department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS, Ames, Iowa).

Molecular serotyping

Molecular serotyping was performed on selected isolates if (i) the isolates were not serotyped by traditional serotyping, (ii) the isolates were classified as untypeable by traditional serotyping, or (iii) serovars predicted by PFGE did not match the serovars reported based on traditional serotyping (Appendix Table 2). Molecular serotyping was performed, as reported by Ranieri et al. [13], using a combination of (i) PCR-based characterization of O antigens and (ii) sequencing-based characterization of the genes encoding the H1 and H2 antigen (i.e., *fli*C and *flj*B, respectively). Briefly, PCR-based characterization of O antigens used a multiplex PCR targeting genes specific for serogroups B, C1, C2-C3, D1, and E1 [14] as well as separate PCRs detecting genes specific for serogroups G and K [15, 16]. *fli*C and *flj*B sequences obtained were aligned and compared to 236 sequences representing 131 *Salmonella* serovars [13] in order to predict H1 and H2 antigens. O antigen PCR and H1 and H2 antigen sequencing results were combined to assign serovar designation consistent with the White-Kauffman-Leminor scheme designations.

Antimicrobial susceptibility testing

Susceptibility testing was performed at Cornell University's Animal Health Diagnostic Center using the Sensititre® system (Trek Diagnostic Systems Ltd., Cleveland, OH). All 1,349 isolates were examined for susceptibility to 15 antimicrobial agents included in the National Antimicrobial Resistance Monitoring System (NARMS) Gram-negative panel: amikacin, amoxicillin/clavulanic acid, ampicillin, cefoxitin, ceftiofur, ceftriaxone, chloramphenicol, ciprofloxacin, gentamicin, kanamycin, nalidixic acid, streptomycin, sulfisoxazole, tetracycline, and trimethoprim/sulfamethoxazole. Clinical and Laboratory Standards Institute guidelines were

used for the interpretation of MIC values when available [17]. Otherwise, MIC values were interpreted using NARMS breakpoints [18].

Pulsed field gel electrophoresis (PFGE)

PFGE typing was performed on 381 representative *Salmonella* isolates. We defined representative isolates as isolates with a unique combination of sampling date, serovar, antimicrobial resistance pattern, farm of origin, and source (i.e., environmental or bovine fecal). If two or more isolates shared a unique combination of the aforementioned criteria, then only one was randomly selected for PFGE typing (using www.random.org) to avoid overrepresentation of subtypes that were re-isolated on the same farm. If a serovar and its variants (e.g., *S.* Typhimurium and *S.* Typhimurium var. O 5 –) were isolated from the same farm on the same date, one representative of each was selected for PFGE typing. PFGE typing was performed using the standard CDC PulseNet protocol for *Salmonella* [19]. Bacterial cultures were embedded in 1% agarose plugs (Lonza SeaKem Gold Agarose, Rockland, ME) and digested with 50 U/plug of *Xba*I (Roche Applied Science, Indianapolis, IN) at 37°C. A subset of *Salmonella* Cerro isolates (n = 10) were also digested with 40 U/plug of *Not*I at 37°C, to improve the resolution [20] of the subtyping results by *Xba*I. The restriction fragments were separated by agarose PFGE using the Chef Mapper® XA or the CHEF-DR II® electrophoresis systems (Bio-Rad, Hercules, California). The initial switch time was 2.16 s and the final switch time was 63.8 s for *Xba*I, and 2 s and 20 s for *Not*I. The gel images were captured using Gel Doc equipment (Bio-Rad, Hercules, California). The tiff images were analyzed using the BioNumerics software program version 5.1 (Applied Maths, Sint-Martens-Latem, Belgium). *Xba*I-PFGE type numbers were assigned after a comparison against 5,828 PFGE types in the BioNumerics database of the Food Safety Laboratory. Clustering analyses were performed using the unweighted pair group method with arithmetic mean algorithm (UPGMA) based on the DICE similarity coefficient with 1.5% position tolerance.

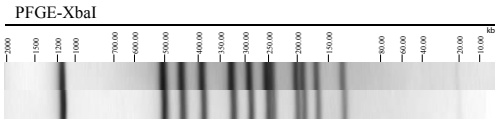
RESULTS

Initial phenotypic subtyping data suggest frequent re-isolation of isolates characterized by a combination of identical serovar and antimicrobial resistance patterns on a given farm.

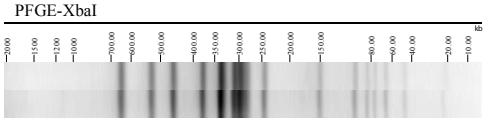
The initial isolate set included 1,349 *Salmonella* isolated from 46 New York dairy farms; 70.2% and 29.8% of isolates were obtained from subclinical dairy cattle and farm environments, respectively (Appendix Table 1). The median herd size for all 46 positive farms was 978 female dairy cattle (range: 245–7,412). Traditional serotyping was performed for 1,344 isolates; ten of these isolates were classified as untypeable by the NVSL (Appendix Table 1). These *Salmonella* isolates were used to initially select 403 representative isolates for PFGE typing; these isolates represented a unique combination of sampling date, serovar, antimicrobial resistance pattern, farm of origin, and source. For example, on farm 1, a total of 8 pansusceptible serovar Meleagridis isolates were obtained from bovine fecal samples on 12/11/2007; one isolate was randomly selected to be included in the set of representative isolates (Appendix Table 1).

Initial PFGE analysis of the 403 representative isolates identified 47 isolates where the PFGE type for a given isolate matched one or more isolates that were classified into a different serovar (Appendix Table 2). For 16 of these 47 isolates, additional isolates with a matching PFGE type were isolated from the same farm as a given isolate. These 16 isolates thus were reclassified as the serovar they were predicted to have, based on PFGE, since the other isolates with the matching PFGE pattern also showed this PFGE predicted serovar. For example, isolate FSL R8-346 from farm 1 was predicted by PFGE to represent serovar Meleagridis but was initially classified as serovar Kentucky by traditional serotyping; as 14 other isolates with the same PFGE isolated from the same farm had been classified as serovar Meleagridis (by classical serotyping), isolate FSL R8-346 was reclassified as Meleagridis (Appendix Table 1). For the other 31 of these 47 isolates, molecular serotyping was performed (see Appendix Table 2 for details). In addition, 10 isolates classified as untypeable and 5 isolates that had not been serotyped were characterized by molecular serotyping (see Appendix Table 2 for details). After completion of molecular serotyping and PFGE analysis, a total of 381 isolates were identified as representative; these isolates were used for the subsequent analyses described below; some of the

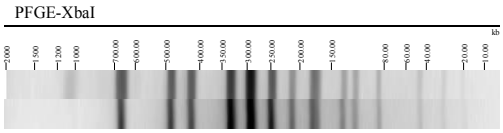
403 isolates that were initially identified as representative were removed from the set of representative isolates as the additional characterization described here revealed that the initial serovar represented a misclassification and that after reclassification this given isolate matched another isolate also included among the isolate set that represented unique subtypes. Interestingly, as part of this analysis we also identified a number of isolates where a given PFGE type represented two different serovars, including (i) PFGE type NYCU.JAAX01.1028, representing serovars Orion 15+ 34+ and Meleagridis; (ii) PFGE type NYCU.JAAX01.0096, representing Kentucky and 8,20:–:z6; and (iii) PFGE type NYCU.JAAX01.0007, representing Muenster and 3,10:–:1,5 (see Figure 2.1 for examples).



FSL No.	Serovar	Source	*Xba*I PFGE Type No.	Farm ID
FSL R8-3193	3,10:-:1,5	Bovine Non-clinical	NYCU.JAAX01.0007	47
FSL R8-4029	Muenster	Environmental	NYCU.JAAX01.0007	62



FSL No.	Serovar	Source	*Xba*I PFGE Type No.	Farm ID
FSL R8-061	8,20:-:z6	Environmental	NYCU.JAAX01.0096	16
FSL R8-065	Kentucky	Bovine Non-clinical	NYCU.JAAX01.0096	26



FSL No.	Serovar	Source	*Xba*I PFGE Type No.	Farm ID
FSL R8-2244	Meleagridis	Environmental	NYCU.JAAX01.1028	11
FSL R8-3428	Orion var.15+ 34+	Environmental	NYCU.JAAX01.1028	64

Figure 2.1 Three instances where *Salmonella* isolates with different serovars shared the same *Xba*I-PFGE type.

More than 50% of representative environmental and subclinical isolates represented serovars Cerro and Kentucky.

Among the 381 representative *Salmonella* isolates, 61.6% (235/381) and 38.3% (146/381) were obtained from the farm environment and subclinical dairy cattle, respectively. These isolates represented 26 serovars (Table 2.1). The 5 most common serovars were Cerro

Table 2.1 Serovar diversity (by source and farm) among the 381 representative *Salmonella* isolates obtained from dairy cattle and farm environments in NY state.

Serovar[a]	Number of environmental isolates	Number of bovine non-clinical isolates	Total Number of isolates	Total Number of farms	Farm ID
Agona	3	—	3	3	10, 28, 46
Anatum	13	7	20	6	4,19,21,39,52,56
Anatum var. 15+	2	2	4	1	39
Cerro	92	56	148	21	15,17,18,26,27,28,30,45,47,49,50, 52,53,54,55,57,59,60,61,62,65
Heidelberg	1	—	1	1	46
Infantis	4	—	4	4	10,18,36,55
Kentucky	36	26	62	10	14,15,16,17,18,19,21,26,42,53
Mbandaka	6	—	6	4	10,40,48,52
Meleagridis	13	6	19	2	1,11
Minnesota	1	3	4	2	49,62
Montevideo	5	1	6	2	41,60
Muenster	5	3	8	5	1,3,23,26,62
Newport	13	16	29	8	14,17,18,19,29,35,60,62
Oranienburg	4	—	4	2	10,19
Orion var. 15+, 34+	2	—	2	1	64
Paratyphi B var. L-tartrate+	1	—	1	1	20
Senftenberg	—	1	1	1	46
Typhimurium	13	14	27	7	1,17,19,22,25,51
Typhimurium var. O 5– (Copenhagen)	6	3	9	4	1,19,25,60
Tennessee	1	—	1	1	46
Thompson	1	—	1	1	57

Table 2.1 (continued)

Serovar[a]	Number of environmental isolates	Number of bovine non-clinical isolates	Total Number of isolates	Total Number of farms	Farm ID
3,10:–:1,5	2	2	4	1	47
3,10:–:l,w	1	—	1	1	1
3,10:e,h:–	1	—	1	1	11
4,5,12:i:–	1	1	2	2	26,52
6,7:–:1,5	1	—	1	1	19
8,20:–:z6	1	—	1	1	16
Untypeable[b]	5	5	10	8	1,15,16,19,39,42,57,65
Total	**235**	**146**	**381**	—	

[a]A total of 26 serovars were found in the 46 farms in this study. Serovar variants (e.g., *S.* Typhimurium and *S.* Typhimurium var. O 5 –) were not considered as individual serovars.

[b]All untypeable isolates were characterized by molecular serotyping, and then classified as Anatum (*n*=2), Cerro (*n*=2), Kentucky (*n*=4), Dublin (n=1), and Meleagridis (*n*=1) (See Appendix Table 2).

(38.8%; 148 isolates obtained from 21 farms); Kentucky (16.3%; 62 isolates from 10 farms); Typhimurium, including variant O 5– (9.4%; 36 isolates from 7 farms); Newport (7.6%; 29 isolates from 8 farms); and Anatum, including variant 15+ (6.3%, 24 isolates from 6 farms). The remaining serovars accounted for 21.5% (82/381). The number of *Salmonella* serovars isolated per farm ranged from 1 to 6 (Table 2.2); for example, farm 19 yielded isolates representing six different serovars, including Typhimurium (including Typhimurium var. O 5–), Kentucky, Newport, Oranienburg, Anatum, and 6,7:–:1,5.

About 25% of environmental and subclinical isolates were resistant to one or more antimicrobial agents, with Salmonella Typhimurium representing the most common drug resistant serovar.

While 76.4% of the 381 isolates designated as representative were pansusceptible, 23.6% (90 isolates) showed resistance to 1 to 11 antimicrobial agents (see Appendix Table 1). These isolates represented 50 different antimicrobial resistance patterns (see Appendix Table 3). All isolates were susceptible to amikacin and ciprofloxacin (Table 2.3). Of the 90 resistant isolates, 42 were isolated from subclinical cattle and 48 were isolated from the farm environment. Among these 90 isolates, the most commonly observed resistances were to ampicillin (found in 72% of these 90 isolates), tetracycline (63% of isolates), and amoxicillin/clavulanic acid (58% of isolates) (Table 2.3). Both pansusceptible as well as resistant *Salmonella* isolates were obtained from 48% (22/46) of the farms; 46% and 6% of farms yielded only pansusceptible or resistant isolates, respectively. Interestingly, in some cases isolates resistant to antimicrobial agents shared the same serovar-*Xba*I-PFGE-type combination as pansusceptible isolates suggesting resistance gene acquisition and/or deletion events that did not affect the PFGE banding pattern.

The 90 isolates that showed resistance to at least one antimicrobial included 14 different serovars, including Typhimurium (25 isolates, representing 20 different resistance patterns), Cerro (13 isolates, representing 12 different resistance patterns), Newport (22 isolates, representing 7 different patterns), Kentucky (8 isolates representing 6 different resistance patterns) (Appendix Table 3).

Table 2.2 Sample collection dates when *Salmonella* positive samples were obtained from each of the 46 farms.

Farm ID	No. of serovars per farm (no. of untypeable isolates)	No. of sample collection dates	Sample collection dates
1	4 (1)	11	10/2/2007–10/22/2008
3	1	1	02/07/2008
4	1	2	07/15/2008–08/18/2008
10	4	4	11/27/2007–09/01/2008
11	2	5	10/31/2007–10/30/2008
14	2	5	10/11/2007–08/30/2008
15	2 (1)	6	10/11/2007–12/09/2008
16	2 (2)	6	12/04/2007–11/11/2008
17	4	9	10/11/2007–09/10/2008
18	4	7	10/18/2007–09/08/2008
19	6 (1)	7	10/18/2007–10/02/2008
20	1	1	06/10/2008
21	2	4	10/19/2007–06/16/2008
22	1	3	12/17/2007–03/14/2008
23	1	1	10/19/2007
25	1	9	10/19/2007–09/03/2008
26	4	9	10/24/2007–10/27/2008
27	1	2	10/24/2007–01/25/2008
28	2	1	10/24/2007
29	1	1	10/24/2007
30	1	18	10/24/2007–10/15/2008
35	1	4	10/23/2008–12/09/2008
36	1	1	11/13/2007
39	1 (1)	5	11/13/2007–12/03/2008
40	1	2	11/14/2007–1/10/2008
41	1	2	08/04/2008–10/03/2008
42	1 (1)	7	11/15/2007–09/17/2008
45	1	1	11/29/2007
46	4	3	11/29/2007–09/25/2008
47	2	3	09/11/2008–01/06/2009
48	1	1	03/2008
49	2	16	01/31/2008–02/23/2009
50	1	4	11/01/2008–02/09/2009
51	1	1	05/09/2008
52	4	6	04/26/2008–02/14/2009
53	2	4	11/06/2008–03/03/2009
54	1	1	11/26/2008

Table 2.2 (continued)

Farm ID	No. of serovars per farm (no. of untypeable isolates)	No. of sample collection dates	Sample collection dates
55	2	3	09/29/2008–03/12/2009
56	1	5	02/22/2008–03/25/2009
57	2 (2)	12	03/19/2008–03/24/2009
59	1	1	09/2008
60	4	7	06/17/2008–04/13/2009
61	1	6	06/17/2008–04/12/2009
62	4	10	06/22/2008–01/22/2009
64	1	2	09/27/2008–02/17/2009
65	1 (1)	10	09/12/2008–08/11/2009

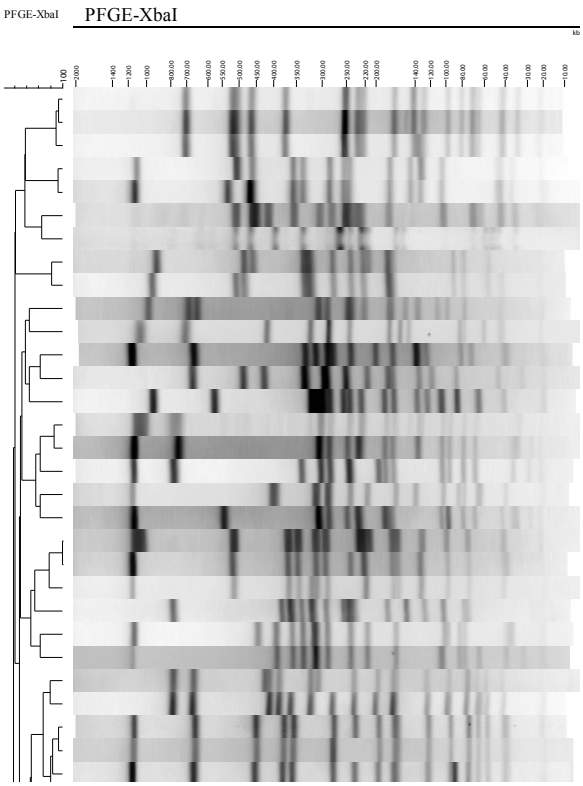
Table 2.3 Antimicrobial resistance observed among the 381 representative *Salmonella* isolated from healthy dairy cattle and associated farm environments in New York.

Antimicrobials	Resistant		Intermediate Resistance	
	Concentration (mg/mL)	No. of isolates	Concentration (mg/mL)	No. of isolates
Amikacin	≥ 64	0	32	0
Amoxicillin/Clavulanic Acid	≥32/16	52	16/8	5
Ampicillin	≥ 32	65	16	2
Cefoxitin	≥ 32	43	16	2
Ceftiofur	≥ 8	48	4	5
Ceftriaxone	≥ 64	5	16–32	28
Chloramphenicol	≥ 32	32	16	13
Ciprofloxacin	≥ 4	0	2	0
Gentamicin	≥ 16	0	8	1
Kanamycin	≥ 64	29	32	0
Nalidixic Acid	≥ 32	1	-	0
Streptomycin	≥ 64	41	-	0
Sulfisoxazole	≥ 512	48	-	0
Tetracycline	≥ 16	57	8	2
Trimethoprim/Sulfamethoxazole	≥ 4/76	2	-	0

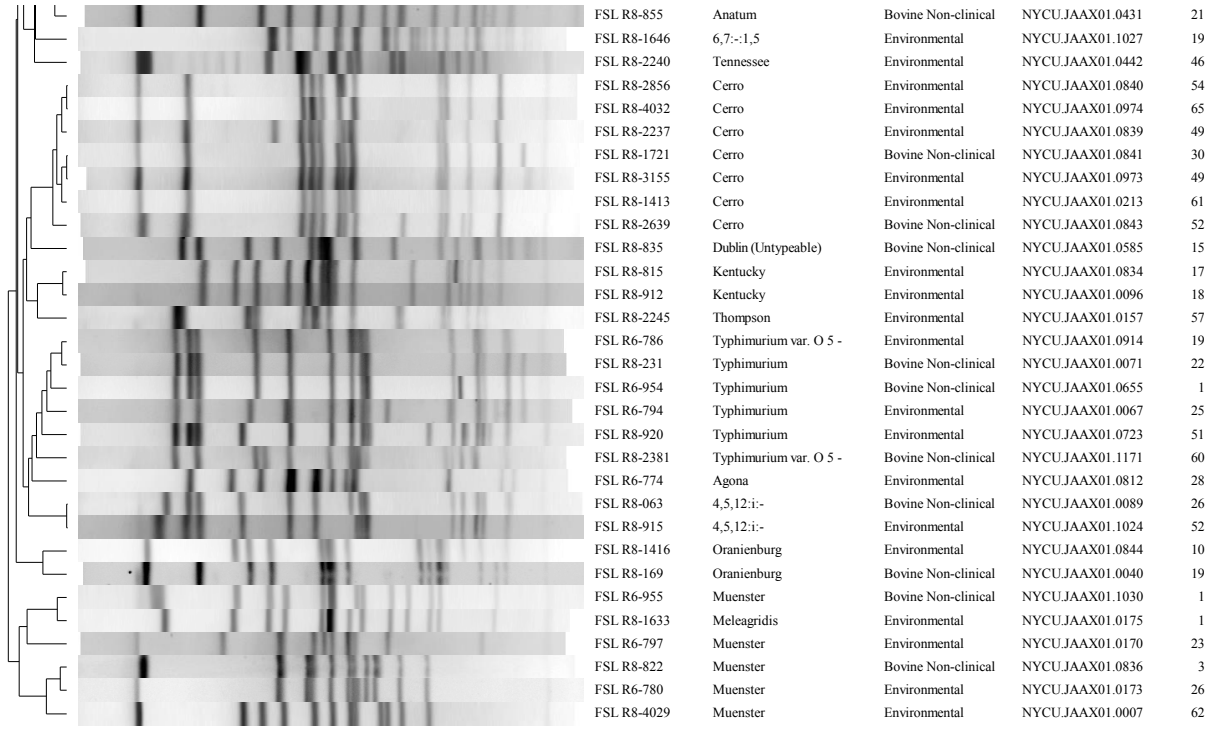
PFGE typing of 381 representative environmental and subclinical isolates revealed presence of PFGE types that exactly match human clinical isolates.

Among the 381 isolates, a total of 61 different *Xba*I-PFGE types were identified (Figure 2.2). On 27 farms, we identified isolates from both environmental samples and fecal samples that shared the same PFGE type (see Figure 2.3 for two examples). *Salmonella* Cerro *Xba*I-PFGE type NYCU.JAAX01.0213 was the most widely distributed PFGE pattern; this pattern represented 134 of the 148 *S.* Cerro isolates (90.5 %), and isolates with this pattern were obtained from 19 farms in 11 different counties in New York. Furthermore, *Not*I-PFGE analyses of 10 randomly chosen serovar Cerro isolates (representing 4 *Xba*I-PFGE types, including 7 isolates with *Xba*I-PFGE type NYCU.JAAX01.0213) yielded two *Not*I-PFGE types with 9 isolates sharing the same *Not*I-PFGE type (Appendix Figure 1).

The 61 *Xba*I-PFGE types identified here were also compared to PFGE types represented among 1,849 human isolates (provided by the New York state Department of Health) in our database; these human isolates were obtained between January 2001 and December 2010, while the 381 isolates characterized here were obtained between October 2007 and August 2009. Human isolates represented a longer time frame to capture a larger number of PFGE types; the goal of this comparison was not to determine whether the farms sampled here were the specific source of human infections but rather to determine whether the PFGE types found among healthy cattle and associated farm environments and human isolates represent overlapping populations. Overall, 14 of the PFGE types represented among the 381 isolates from dairy farms exactly matched PFGE types found among human isolates. These PFGE patterns represent 12 different serovars (Table 2.4). A total of 143 human isolates (7.7% of the 1,849 human isolates) were represented by PFGE types that were identical to PFGE types found among *Salmonella* isolates from dairy farms (Table 2.4).

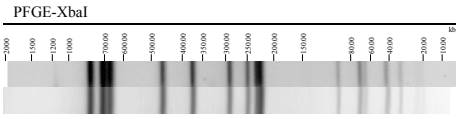


FSL No.	Serovar	Source	*Xba*I-PFGE Type No.	Farm ID
FSL R8-1623	Typhimurium var. O 5 -	Environmental	NYCU.JAAX01.0104	25
FSL R8-1642	Typhimurium var. O 5 -	Environmental	NYCU.JAAX01.1031	25
FSL R8-1622	Typhimurium	Environmental	NYCU.JAAX01.1034	25
FSL R8-2014	Mbandaka	Environmental	NYCU.JAAX01.0757	10
FSL R8-2007	Mbandaka	Environmental	NYCU.JAAX01.0758	52
FSL R8-2241	Heidelberg	Environmental	NYCU.JAAX01.1016	46
FSL R8-900	Kentucky	Environmental	NYCU.JAAX01.0184	16
FSL R8-2648	Minnesota	Bovine Non-clinical	NYCU.JAAX01.0523	62
FSL R8-2694	Minnesota	Bovine Non-clinical	NYCU.JAAX01.1029	49
FSL R8-1403	Paratyphi B var. L- tartrate+	Environmental	NYCU.JAAX01.0187	20
FSL R8-821	Senftenberg	Bovine Non-clinical	NYCU.JAAX01.0372	46
FSL R8-2812	Montevideo	Environmental	NYCU.JAAX01.0156	41
FSL R8-3428	Orion var. 15+ 34+	Environmental	NYCU.JAAX01.1028	64
FSL R8-3417	Montevideo	Environmental	NYCU.JAAX01.1006	60
FSL R8-2249	Newport	Environmental	NYCU.JAAX01.1032	35
FSL R8-3136	Newport	Environmental	NYCU.JAAX01.0126	35
FSL R6-961	Newport	Bovine Non-clinical	NYCU.JAAX01.0328	18
FSL R6-932	Infantis	Environmental	NYCU.JAAX01.0114	36
FSL R8-2900	Newport	Bovine Non-clinical	NYCU.JAAX01.0121	62
FSL R6-965	Newport	Bovine Non-clinical	NYCU.JAAX01.1015	19
FSL R8-2649	Newport	Bovine Non-clinical	NYCU.JAAX01.1014	60
FSL R8-1683	Newport	Bovine Non-clinical	NYCU.JAAX01.1017	60
FSL R8-060	Agona	Environmental	NYCU.JAAX01.0807	46
FSL R8-1392	Infantis	Environmental	NYCU.JAAX01.0261	10
FSL R6-791	Infantis	Environmental	NYCU.JAAX01.0107	18
FSL R8-2634	Anatum	Bovine Non-clinical	NYCU.JAAX01.1026	4
FSL R8-350	Anatum	Environmental	NYCU.JAAX01.0430	56
FSL R6-928	Anatum	Environmental	NYCU.JAAX01.0424	39
FSL R6-929	Anatum var. 15+	Environmental	NYCU.JAAX01.0423	39
FSL R8-264	Anatum	Bovine Non-clinical	NYCU.JAAX01.1025	39

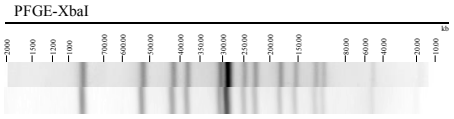


FSL R8-855	Anatum	Bovine Non-clinical	NYCU.JAAX01.0431	21
FSL R8-1646	6,7:-:1,5	Environmental	NYCU.JAAX01.1027	19
FSL R8-2240	Tennessee	Environmental	NYCU.JAAX01.0442	46
FSL R8-2856	Cerro	Environmental	NYCU.JAAX01.0840	54
FSL R8-4032	Cerro	Environmental	NYCU.JAAX01.0974	65
FSL R8-2237	Cerro	Environmental	NYCU.JAAX01.0839	49
FSL R8-1721	Cerro	Bovine Non-clinical	NYCU.JAAX01.0841	30
FSL R8-3155	Cerro	Environmental	NYCU.JAAX01.0973	49
FSL R8-1413	Cerro	Environmental	NYCU.JAAX01.0213	61
FSL R8-2639	Cerro	Bovine Non-clinical	NYCU.JAAX01.0843	52
FSL R8-835	Dublin (Untypeable)	Bovine Non-clinical	NYCU.JAAX01.0585	15
FSL R8-815	Kentucky	Environmental	NYCU.JAAX01.0834	17
FSL R8-912	Kentucky	Environmental	NYCU.JAAX01.0096	18
FSL R8-2245	Thompson	Environmental	NYCU.JAAX01.0157	57
FSL R6-786	Typhimurium var. O 5 -	Environmental	NYCU.JAAX01.0914	19
FSL R8-231	Typhimurium	Bovine Non-clinical	NYCU.JAAX01.0071	22
FSL R6-954	Typhimurium	Bovine Non-clinical	NYCU.JAAX01.0655	1
FSL R6-794	Typhimurium	Environmental	NYCU.JAAX01.0067	25
FSL R8-920	Typhimurium	Environmental	NYCU.JAAX01.0723	51
FSL R8-2381	Typhimurium var. O 5 -	Bovine Non-clinical	NYCU.JAAX01.1171	60
FSL R6-774	Agona	Environmental	NYCU.JAAX01.0812	28
FSL R8-063	4,5,12:i:-	Bovine Non-clinical	NYCU.JAAX01.0089	26
FSL R8-915	4,5,12:i:-	Environmental	NYCU.JAAX01.1024	52
FSL R8-1416	Oranienburg	Environmental	NYCU.JAAX01.0844	10
FSL R8-169	Oranienburg	Bovine Non-clinical	NYCU.JAAX01.0040	19
FSL R6-955	Muenster	Bovine Non-clinical	NYCU.JAAX01.1030	1
FSL R8-1633	Meleagridis	Environmental	NYCU.JAAX01.0175	1
FSL R6-797	Muenster	Environmental	NYCU.JAAX01.0170	23
FSL R8-822	Muenster	Bovine Non-clinical	NYCU.JAAX01.0836	3
FSL R6-780	Muenster	Environmental	NYCU.JAAX01.0173	26
FSL R8-4029	Muenster	Environmental	NYCU.JAAX01.0007	62

Figure 2.2 The 61 *Xba*I-PFGE types found among the 381 representative *Salmonella* isolates characterized here. These isolates were obtained from subclinical dairy cattle and associated dairy farm environments samples collected in 46 New York state dairy farms.



FSL No.	Serovar	Source	*Xba*I PFGE Type No.	Farm ID
FSL R8-171	Typhimurium	Environmental	NYCU.JAAX01.0071	22
FSL R8-271	Typhimurium	Bovine Non-clinical	NYCU.JAAX01.0071	22



FSL No.	Serovar	Source	*Xba*I PFGE Type No.	Farm ID
FSL R6-810	Meleagridis	Bovine Non-clinical	NYCU.JAAX01.0175	1
FSL R8-1406	Meleagridis	Environmental	NYCU.JAAX01.0175	1

Figure 2.3 Examples where *Salmonella* isolated from different sources within the same farm shared undistinguishable *Xba*I-PFGE types. Shown here are environmental and fecal isolates from Farms 1 and 22 that shared the same PFGE type; overall (including these two farms), there were 27 farms where isolates from different sources within the same farm shared undistinguishable *Xba*I-PFGE types.

Table 2.4 PFGE types found among both, isolates from subclinical dairy cattle and farm environments, (collected between October 2007 and August 2009) and of isolates from human clinical cases (reported to the New York State Department of Health between January 2001 and December 2010).

PFGE type	Serovar	No. of human isolates	No. of isolates obtained from farm environments and subclinical animals
NYCU.JAAX01.0040	Oranienburg	13	1
NYCU.JAAX01.0071	Typhimurium	31	14
NYCU.JAAX01.0089	4,5,12:i:–	31	1
NYCU.JAAX01.0096	Kentucky	1	66
NYCU.JAAX01.0114	Infantis	1	1
NYCU.JAAX01.0121	Newport	4	15
NYCU.JAAX01.0126	Newport	8	8
NYCU.JAAX01.0156	Montevideo	11	3
NYCU.JAAX01.0157	Thompson	31	1
NYCU.JAAX01.0213	Cerro	3	136
NYCU.JAAX01.0442	Tennessee	5	1
NYCU.JAAX01.0757	Mbandaka	2	1
NYCU.JAAX01.0914	Typhimurium	1	2
NYCU.JAAX01.1016	Heidelberg	1	1

DISCUSSION

Salmonella serovars Cerro and Kentucky were the most common serovars isolated from subclinical dairy cattle and dairy farm environments in New York.

Salmonella serovars Cerro and Kentucky were the two most common serovars isolated from subclinical dairy cattle and dairy farm environments. While farms that yielded *Salmonella* isolates on multiple sample collection dates could drive serovar and subtype frequencies (as isolates with a given serovar may have been included for multiple sampling dates from a given farm), re-isolation of a serovar also makes exposure and dispersal of this serovar more likely. Our data are consistent with the NAHMS Dairy 2007 study, which found serovars Cerro and Kentucky to be the most common serovars isolated from healthy cattle on U.S. dairy operations; in this study, 121 dairy farms were sampled in 17 states that host 79.5% of U.S. dairy herds and 82.5% of U.S. dairy cows [10]. *Salmonella* Cerro was also the most common serovar found in bulk tank milk and milk filters tested as part of the NAHMS study [21]. van Kessel et al. [22] previously suggested that *S.* Cerro might behave as a commensal organism in dairy cattle, based on a protracted outbreak of subclinical *S.* Cerro infection. However, *S.* Cerro has been described as an emerging pathogen among dairy cattle in the northeastern U.S. over the last few years, as evidenced by a sharp rise in *S.* Cerro isolations from bovine clinical cases [11, 23, 24]. These studies have also reported that, based on PFGE analysis, U.S. *Salmonella* Cerro isolates represent a highly clonal population, consistent with our PFGE data reported here. While our data support frequent isolation of *S.* Cerro from cattle that are subclinically infected, experimental infection studies will be needed to further characterize this serovar's behavior as a commensal vs. pathogenic organism.

In addition to serovars Cerro and Kentucky, a number of other serovars commonly isolated from the New York dairy farms in our study, were also reported by the 2007 NAHMS study [10] as commonly isolated from dairy operations and healthy cattle around the U.S. (e.g., Muenster, Meleagridis, and Mbandaka). Interestingly, none of these serovars (i.e., Cerro, Kentucky, Muenster, Meleagridis, and Mbandaka) are among the 20 most common human

disease-associated serovars reported by CDC [25]. These findings suggest that a number of *Salmonella* serovars that are common among subclinically infected cattle are rare among human clinical cases. Future phenotypic and genotypic studies on these serovars may be warranted to identify possible mechanisms that may explain an association of these serovars with bovine hosts.

Serovars resulting from traditional serotyping could be confirmed using a combination of PFGE typing and molecular serotyping.

PFGE typing has previously been demonstrated to be useful in serovar prediction [26], particularly if large databases are available to facilitate serovar prediction [13]. We thus used the PFGE data for our isolates to determine whether serovar identification by classical serotyping matched the serovar predicted based on a comparison of PFGE types to a database of previously characterized isolates with both PFGE and classical serovar data. Here, this approach successfully identified a number of isolates that did not have the correct serovar assigned to them. We also identified a number of instances where different serovars with similar antigenic formula were found to have indistinguishable PFGE types, such as Kentucky (8,20:i:z6) and 8,20:–:z6, among others. This is consistent with previous reports [27, 28] that also identified isolates, with similar antigenic formula, that shared indistinguishable PFGE types. These findings support that prediction of serovar using banding pattern-based subtyping methods is likely to yield at least some inconsistent results. In contrast, use of a molecular serotyping approach based on PCR amplification of O-antigen specific genes and sequencing of *fliC* and *fljB* [13] provided good differentiation of isolates and clarification of ambiguous traditional serotyping results (e.g., where traditional serotyping data and PFGE-based prediction of serovars did not match), as well as for improved classification of untypeable isolates. These findings suggest that the previously described [13] molecular serotyping approach used here provides a suitable tool for the assessment of *Salmonella* serovars, particularly as PCR reactions that identify additional O antigens are developed.

Some Salmonella serovars and subtypes frequently recovered from human clinical cases were also regularly found among healthy cattle and dairy farm environments.

In addition to *Salmonella* serovars that were frequently isolated from healthy cattle and dairy farm environments but rare among human clinical cases, we also identified a number of serovars commonly associated with human clinical cases. For example, the 3rd and 4th most commonly isolated serovars in this study were Typhimurium and Newport, which are among the top 3 *Salmonella* serovars isolated from laboratory-confirmed human cases in the U.S. [25]. Furthermore, 10 of the 26 serovars isolated here were reported among the 20 most commonly isolated serovars from human clinical cases in 2009 [25]. When the PFGE types for 381 isolates from subclinical cattle and farm environments were compared to 1,849 PFGE types from human isolates collected in New York, a total of 14 PFGE types representing 12 serovars were indistinguishable from PFGE types from human isolates. Even though *Salmonella* Cerro is rarely associated with human illness, three human isolates matched Cerro isolates from healthy cattle and dairy farm environments by PFGE, indicating that Cerro may, in rare cases, cause human illnesses, consistent with a few reports of *Salmonella* Cerro-associated human salmonellosis [24, 29, 30]. While our data do not specifically show that the farms sampled here were sources of human cases, our data support that subclinically infected cattle as well as farm environments may represent reservoirs or sources of *Salmonella* serovars and PFGE types commonly associated with human disease. Similarly, other reports have shown that a considerable proportion of *Salmonella* isolates from dairy cattle with clinical signs can match human clinical isolates by PFGE [31].

Subclinical cattle and dairy farm environments are sources of drug-resistant Salmonella.

Our data also showed that a number of *Salmonella* isolates from healthy cattle and farm environments are resistant to antimicrobial drugs, including some isolates resistant to 8 or more drugs. Identification of a number of MDR *S.* Newport is consistent with previous studies that found a considerable number of MDR *S.* Newport among cattle with clinical salmonellosis [32, 33]. Previous studies have also found antimicrobial drug-resistant *Salmonella*, including MDR

isolates, in healthy cattle and dairy farms in Thailand and the U.S. [34, 35]. In another study, Perron et al. [9] showed that *Salmonella* from subclinically infected livestock representing species other than cattle (i.e., swine) can also be drug-resistant. Overall, these data suggest that not only clinically affected animals, but also healthy animals and farm environments not associated with animal salmonellosis cases or outbreaks, can be possible sources of MDR *Salmonella*.

CONCLUSION

Our data indicate that healthy livestock and farm environments may represent a potentially important reservoir of *Salmonella* serovars and subtypes associated with human infections, particularly considering that nearly 25% of *Salmonella* isolates were resistant to multiple antimicrobial agents. Subclinical shedding of *Salmonella* by dairy cattle thus represents a potential public health issue, particularly because fecal shedding results in widespread environmental contamination and an increased risk of within-herd transmission, both of which can promote zoonotic transmission through foodborne exposure or direct contact (e.g., for farm visitors). In addition, shedding animals cannot be recognized through clinical signs, which reduces the likelihood of adequate biosecurity efforts and quarantine efforts for these non-clinical shedders. While some studies have suggested that direct contact with cattle or other livestock is a risk factor for acquiring human salmonellosis [36], further studies will be needed to quantitatively define the risk of transmission from healthy dairy cattle or cattle with clinical signs of salmonellosis and dairy-associated environments to humans.

ACKNOWLEDGEMENTS

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CHAPTER 3

GENOMIC CHARACTERIZATION OF *SALMONELLA* CERRO ST367, AN EMERGING *SALMONELLA* SUBTYPE IN CATTLE IN THE UNITED STATES*

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ABSTRACT

Within the last decade, *Salmonella enterica* subsp. *enterica* serovar Cerro (*S.* Cerro) has become one of the most common serovars isolated from cattle and dairy farm environments in the northeastern US. The fact that this serovar is commonly isolated from subclinically infected cattle and is rarely associated with human disease, despite its frequent isolation from cattle, has led to the hypothesis that this emerging serovar may be characterized by reduced virulence. We applied comparative and population genomic approaches to (i) characterize the evolution of this recently emerged serovar and to (ii) gain a better understanding of genomic features that could explain some of the unique epidemiological features associated with this serovar. In addition to generating a *de novo* draft genome for one *Salmonella* Cerro strain, we also generated whole genome sequence data for 26 additional *S.* Cerro isolates, including 16 from cattle operations in New York (NY) state, 2 from human clinical cases from NY in 2008, and 8 from diverse animal sources (7 from Washington state and 1 from Florida). All isolates sequenced in this study represent sequence type ST367. Population genomic analysis showed that isolates from the NY cattle operations form a well-supported clade within *S.* Cerro ST367 (designated here "NY bovine clade"), distinct from isolates from Washington state, Florida and the human clinical cases. A molecular clock analysis indicates that the most recent common ancestor of the NY bovine clade dates back to 1998, supporting the recent emergence of this clone.

Comparative genomic analyses revealed several relevant genomic features of *S.* Cerro ST367, that may be responsible for reduced virulence of *S.* Cerro, including an insertion creating a premature stop codon in *sopA*. In addition, patterns of gene deletion in *S.* Cerro ST367 further support adaptation of this clone to a unique ecological or host related niche. Our results indicate that the increase in prevalence of *S.* Cerro ST367 is caused by a highly clonal subpopulation and that *S.* Cerro ST367 is characterized by unique genomic deletions that may indicate adaptation to specific ecological niches and possibly reduced virulence in some hosts.

INTRODUCTION

Genomic characteristics associated with the emergence or reemergence of pathogens in livestock operations can be subdivided into two categories; (i) genomic features that increase the adaptation to a host, or facilitate the jump to a new host species, or (ii) genomic features that provide increased adaptation to environmental factors in the livestock environment, such as antibiotic resistance. Comparative and population genomic studies are particularly suited to determine which features are responsible for the emergence of certain pathogens. For instance, Price et al. [1] showed that a putative host jump, from humans to livestock, in a clonal complex in *Staphylococcus aureus* was associated with the loss of phage-carried human virulence genes and with the acquisition of tetracycline and methicillin resistance.

Salmonella enterica is one of the most frequent causes of bacterial foodborne illness and death in the United States [1, 2]. In *Salmonella*, examples of emergent clones include *S.* Typhimurium DT 104, a multidrug resistant clone, which has seen a global epidemic spread from 1990 [3], and *S. enterica* serovar 4,5,12:i:–, a monophasic variant of *S.* Typhimurium, which showed a global increase in the mid-1990s [4]. In this study, we present comparative and population genomic research on *S. enterica* subsp. *enterica* serovar Cerro (*S.* Cerro). *S.* Cerro is rarely associated with human disease, with only one outbreak reported in the US so far that could be solely attributed to this serovar [5]; an additional outbreak was recently reported and it was linked to multiple serovars, including *S.* Cerro [6]. However, this *Salmonella* serovar has emerged over the last decade as one of the most abundant *Salmonella* serovars in cattle operations in the northeastern US [7], including one of the most common serovars among asymptomatic dairy cattle and in the dairy farm environment [8] in the northeastern United States. Most of the *S.* Cerro isolated from cattle and farms represent one pulsed field electrophoresis (PFGE) type, indicating that a single clonal lineage is involved in this emergence [7]. It is unknown what causes *S.* Cerro to be associated with cattle and why it is rarely involved in human disease. Therefore, we hypothesize that *S.* Cerro has distinct genomic characteristics that explain its association with cattle and limited association with human disease.

METHODS

Isolates selection

The 27 *S.* Cerro isolates for genome sequencing (*n=1*) and re-sequencing (*n=26*) were isolated from 1986 to 2008 from human cases and domesticated and wild animals in 3 different states (i.e., New York, Washington, and Florida; Table 3.1).

Table 3.1 27 *Salmonella* Cerro isolates sequenced in this study

FSL No.[a]	Source	Date of isolation	Obtained from[b]	County and/or State of origin	SRA accession[c]
R8-4199	Canine host	Oct-1989	WSU	WA	SRR654177
R8-4201	Feline host	Jun-1990	WSU	WA	SRR654178
R8-4194	Feline host	Dec-1986	WSU	FL	SRR654174
R8-4196	Bovine host	Jul-1987	WSU	Grant, WA	SRR654176
R8-4235	Bovine host	Aug-2001	WSU	Yakima, WA	SRR654180
R8-4285	Bovine host	Aug-2007	WSU	Yakima, WA	SRR654183
R8-4271	Bovine host	Jan-2006	WSU	Grant, WA	SRR654182
R8-4204	Bovine host	Jan-2000	WSU	Yakima, WA	SRR654179
R8-3973	Human host	2008	NYSDOH	NY	SRR653053
R8-3972*	Human host	2007	NYSDOH	NY	SRR653052
R8-2827*	Farm Environment	Oct-2008	CU-Warnick	Tompkins, NY	SRR653002
R8-2660*	Bovine host, non-clinical	Sep-2008	CU-Warnick	Niagara, NY	SRR654036
R8-2280*	Bovine host, clinical	Apr-2008	CAHDC	Wyoming, NY	SRR653929
R8-0257	Bovine host, non-clinical	Jan-2008	CU-Warnick	Genesee, NY	SRR653610
R8-1413	Farm Environment	Jun-2008	CU-Warnick	Niagara, NY	SRR653005
R8-1441	Bovine host, non-nlinical	May-2008	CU-Warnick	Steuben, NY	SRR653928
R8-0358	Bovine host, non-clinical	Jan-2008	CU-Warnick	Steuben, NY	SRR653721
R8-0245	Bovine host, non-clinical	Jan-2008	CU-Warnick	Genesee, NY	SRR653609
R8-2349	Bovine host, clinical	Jun-2008	CAHDC	Livingston, NY	SRR653931
R8-1415	Farm Environment	Jun-2008	CU-Warnick	Niagara, NY	SRR653010
R8-2008	Farm Environment	Aug-2008	CU-Warnick	Franklin, NY	SRR653009
R8-2639	Bovine host, non-clinical	Aug-2008	CU-Warnick	Franklin, NY	SRR654035
R8-3258	Bovine host, clinical	Jul-2008	CAHDC	Livingston, NY	SRR654173
R8-1044	Bovine host, non-clinical	Apr-2008	CU-Warnick	Genesee, NY	SRR653927
R8-2237	Farm Environment	Sep-2008	CU-Warnick	Steuben, NY	SRR652998
R8-0235	Bovine host, non-clinical	Jan-2008	CU-Warnick	Wyoming, NY	SRR654552
R8-1390	Farm Environment	May-2008	CU-Warnick	Steuben, NY	SRR652996

[a] *Isolates marked with an asterisk were used in the Caco-2 invasion assays.*

[b] WSU = Washington State University; NYSDOH = New York State Department of Health; CU-Warnick = Cornell University, Warnick laboratory; CAHDC = *Animal Health Diagnostic Center, Cornell University.*
[c] *SRA = Sequence Read Archive (www.ncbi.nlm.nih.gov/sra).*

Genome sequencing, assembly and annotation

The genome of *S.* Cerro FSL R8-0235 was sequenced using the SOLiD™ system (Applied Biosystems, Foster City). Mate-paired 50 bp reads were obtained and a *de novo* assembly was performed as detailed in Den Bakker et al. [24]. Contigs longer than 200 bp were submitted to the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP) [43] for automated annotation. Unpaired 50 bp reads for the genomes of the additional 26 *S.* Cerro ST367 isolates were obtained using the SOLiD™ system (Applied Biosystems, Foster City) as detailed in Den Bakker et al. [44].

Prophage identification

PROPHINDER [11] was used to find putative prophages. The prophage regions were compared, using RAST [12], to previously sequenced genomes to identify homologous regions.

SOLiD read mapping, population genetics analysis, and read mapping based gene presence/absence analysis

SOLiD reads were mapped against a reference genome (FSL R8_0235) using PerM [45]. ComB [46] was used to for SNP calling and creation of consensus sequences. Regions with coverage less than 10X were masked in the consensus sequences. Consensus sequences created with ComB were used as input for the BratNextGen [19] recombination detection software, using 100 replicates of 50 iterations each. SNPs in regions that were predicted to be involved in a recombination event with $P < 0.01$ were excluded from the analysis.

A maximum likelihood (ML) tree based on the SNP data was created in MEGA 5 [47], and this ML tree was used to test for the presence of a temporal signal in the dataset using Path-O-Gen 1.4 (available from http://tree.bio.ed.ac.uk/software/pathogen/). BEAST version 1.7.5 [48] was used to create a tip-dated phylogeny of the *S.* Cerro isolates. Four different models differing in assumptions on mutation rate and effective population size (strict clock, constant

population size; strict clock, Gaussian Markov random field (GMRF) model [49]; relaxed clock, constant population size; relaxed clock, GMRF model) were ran for 10 million generations each and compared using the Bayes factor as implemented in Tracer version 1.5 (A. Rambaut available from http://tree.bio.ed.ac.uk/software/tracer/).

Read mapping based gene presence/absence analysis was performed by mapping SOLiD reads to selected reference genomes using PerM [45]. Coverage per annotated gene feature in the reference genome was subsequently obtained using the 'coverage' tool from the BEDtools suite [50].

Caco-2 cell invasion assays of S. Cerro, S. Kentucky, S. Typhimurium, and S. Newport

To compare the ability of *S.* Cerro isolates to invade human intestinal epithelial cells, Caco-2 cells were infected with *S.* Typhimurium ($n = 4$), *S.* Newport ($n = 4$), *S.* Kentucky ($n = 4$), and *S.* Cerro ($n = 4$) (Appendix Table 4). *Salmonella* Typhimurium ATCC® 14028 was used as a positive control and its *sir*A isogenic mutant as a negative control. All isolates were susceptible to gentamicin as determined by antimicrobial susceptibility testing (MIC values between 0.25 and 1 μg/ml) by the Cornell University Animal Health Diagnostic Center. *Salmonella* isolates were grown on Luria Bertani (LB) plates at 37°C for 16 hours. A colony was transferred into 5 mL LB broth and incubated 18 hours at 37°C, without shaking. After 18 hours of incubation, 1 mL of each culture was pelleted by centrifugation and re-suspended in 1 mL of Phosphate Buffered Saline (PBS) pH 7.4. Bacterial cells were diluted and Caco-2 cells were inoculated at an MOI of 10. Each strain was inoculated in triplicate in each of the 3 experiments conducted. Appropriate dilutions were plated on LB for calculation of the initial inoculum.

For all the experiments Caco-2 cells were maintained in Dulbecco's Modified Eagle Medium (DMEM) 20% FBS 1% non-essential amino acids at 37°C and 5.0% CO_2, for no more than 50 passages. The 24-well plates were seeded at a concentration of 5.0×10^4 cells/well and incubated at 37°C and 5% CO_2 for 48 hours. Thirty minutes before the cells were inoculated with *Salmonella*, media in the 24-well plate was replaced with fresh media. Caco-2 cells were inoculated, and incubated at 37°C and 5% CO_2 for 1 hour, followed by 3 washes with pre-

warmed PBS. Fresh media was distributed into each well followed by a 15 minute incubation at 37°C and 5% CO_2. Finally, media with gentamicin (50 μg/mL) was added and the cells were incubated for 1 hour at 37°C and 5% CO_2. The cells were then lysed by vigorously pipetting 500 μL of chilled water in each well. The bacterial suspensions recovered were plated on LB and incubated at 37°C overnight. Invasion efficiency was calculated as [CFU recovered/CFU infected]×100. Statistical analysis was performed using SAS software (SAS Institute Inc., Cary, NC, USA). The invasion efficiencies were analyzed using one-way analysis of variance (ANOVA), Tukey post hoc test, and the data was log-transformed to satisfy ANOVA assumptions of normality.

RESULTS AND DISCUSSION

De novo assembly shows that S. Cerro FSL R8-0235 has a genome size of approximately 4.7 Mbp, contains six prophage regions and represents MLST sequence type ST367.

After exclusion of contigs fewer than 200 bp, the total length of the *S.* Cerro FSL R8-0235 draft *de novo* assembly was 4,675,817 bp. The assembly consisted of 126 contigs, with a contig N50 of 292,947 bp, and a maximum contig length of 691,181 bp. The average coverage depth of the assembly was 96X. One contig, contig 016, contained genes of an IncI1-like plasmid, however it is unclear whether this is an integrated or extrachromosomal plasmid. In addition to genes involved in plasmid transfer, stability and replication, this plasmid also carries genes encoding a resistance nodulation division (RND) efflux pump [9]. However, none of the isolates sequenced in this study showed resistance to single or multiple antimicrobial agents. No evidence for the existence of additional plasmids within the genome was found. This may be at least partially due to the presence of a DNA phosphorothioation-dependent restriction modification (RM) system in all *S.* Cerro strains examined in this study. While this RM system has been well characterized in *S.* Cerro [10] , a PSI-BLAST search reveals this type of RM system is very rare among *Salmonella*, and only found in a limited number of sequenced

Salmonella strains of serovars Saintpaul (SARA23, str. 9712, str. JO2008), Namur (str.05-2929) and Panama (ATCC 7378).

Prediction of lysogenic prophages and prophage remnants in the *S.* Cerro FSL R8-0235 genome was performed using Prophinder [11]. Six putative prophages or remnants of prophages, ranging in length from 5.78 to 31.52 Kb, were predicted to be present in the *S.* Cerro FSL R8-0235 genome (Table 3.2). The six prophage regions, which we refer to as prophage 57014, 57017, 57018, 57023, 57024, and 57025, were compared, using RAST [12], to previously sequenced genomes to identify homologous regions. Prophages 57023 and 57025 (Table 3.2) are similar in composition to a *S.* Typhimurium ES18-like bacteriophage, while 57014 shows similarity to an Enterobacteria P22-like prophage. While typical *Salmonella*-associated prophages, such as Gifsy-1, Gifsy-2, Fels-1, and Fels-2 [13-15] were not predicted to be present in the *S.* Cerro FSL R8-0235 genome, prophage 57024 shared many genes with a prophage found in *Photorhabdus luminescens* subsp. *laumondii* TT01, which has been described as a successful insect pathogen as well as symbiont of soil entomopathogenic nematodes [16].

Genome assembly based multi locus sequence typing (MLST) was performed using the online tool [17] of the Center for Genomic Epidemiology (Lyngby, Denmark; http://www.genomicepidemiology.org/) and an additional BLASTN search. This analysis revealed that *S.* Cerro FSL R8-0235 belongs to sequence type (ST) 367. According to the *Salmonella* MLST database (http://mlst.ucc.ie/mlst/dbs/Senterica) ST367 is associated with a *S.* Cerro isolate from a human case in Germany in 1985. The database also contains an accession of the type strain of *S.* Cerro, isolated from swine in 1936 in Uruguay. This strain belongs to ST1291 and displays a different allelic type at each of the seven MLST loci. *S.* Cerro therefore is very likely to be polyphyletic, which makes interpretation of historical references without genomic or MLST sequence data difficult. Because all isolates sequenced in this study belong to ST367, we will refer to these isolates as *S.* Cerro ST367 from here on. Timme et al. [18] recently published sequence data for another *S.* Cerro ST367 strain (strain 818; NZ_AOZJ00000000); this group showed that, among all serovars that have been sequenced so far, *S.* Adelaide FSL A4-669

Table 3.2 Prophage distribution in the *Salmonella* Cerro FSL R8-0235 genome.

Prophage ID	Contig[a]	Length	Previously described phages similar to *S.* Cerro prophages
57018	003	5,780 bp	Putative prophage remnant, found in *E. coli*, *S.* Typhi (CT18), *S.* Newport (SL254).
57024	009	2,7456 bp	Similar to prophage in *S.* Baildon (FSL R6-199) as well as prophages in *E. coli* and *Photorhabdus luminescens* subsp. *laumondii* TT01
57025	009	31,520 bp	*S.* Typhimurium bacteriophage ES18-like, similar prophages in *S.* Senftenberg (FSL A4-543), *S.* Schwarzengrund (CVM19633), and *S.* Montevideo (FSL S5-403)
57023	013	15,396 bp	*S.* Typhimurium bacteriophage ES18-like, similar prophages in *S.* Senftenberg (FSL A4-543) and *S.* Choleraesuis (SC-B67)
57017	017	7,296 bp	Putative prophage remnant, found in a wide variety of *Salmonella enterica* serovars, *E. coli* and *Shigella*
57014	018	11,952 bp	Enterobacteria P22 phage, similar prophages are found in *S.* Dublin (CT_02021853), *S.* Paratyphi A (ATCC 9150, AKU_12601)

[a]Contig in the *S.* Cerro pseudogenome where the predicted prophage is encoded.

is most closely related to *S.* Cerro ST367 which is consistent with our study (see below).

Population genomic analysis of 27 Salmonella serovar Cerro isolates suggests a recent clonal expansion of a bovine-associated S. Cerro lineage.

To infer whether the *S.* Cerro isolates associated with bovine hosts and cattle-associated environments form separate subpopulations from *S.* Cerro isolated from other sources, we obtained whole genome sequencing data for 26 additional isolates (Table 3.1). After removal of putative recombinogenic regions, as identified by BratNextGen [19], and SNPs that were present in fewer than 90% of the isolates, 343 SNPs were left for analysis. To assess the presence of a temporal signal in the dataset, a Path-O-Gen (available from http://tree.bio.ed.ac.uk/software/pathogen/) analysis was performed using a maximum likelihood tree inferred from the SNP data set. This analysis showed a correlation ($R^2 = 0.645$) between the time of isolation of the individual isolates and the root-to-tip divergence, indicating a temporal signal for this dataset and justifying a molecular clock based phylogenetic analysis. A Bayesian analysis, assuming a relaxed molecular clock and a constant population size, inferred the mean mutation rate for the core genome of the 27 *S.* Cerro isolates to be 2.4×10^{-7}/site/year (95% Highest Probability Density (HPD) $1.5 \times 10^{-7} – 3.3 \times 10^{-7}$). This mutation rate is comparable to mutation rates estimated for *Buchnera aphidicola* [20] and *Helicobacter pylori* [21], but about twice as fast as recently inferred for *S.* Agona [22]. The New York bovine isolates are found in a well-supported (posterior probability 1.0) clade (NY bovine clade; see Figure 3.1), well separated from the isolates from Washington state, Florida, and the human clinical isolates from New York state. This may indicate that, although isolates of *S.* Cerro of the bovine-associated clade were prevalent in farm environments, and thus farm personnel would be frequently exposed to this clone, this clone was not responsible for the human cases in New York state represented by these two isolates. The time of the emergence of the most recent ancestor (MRCA) of the NY bovine clade is estimated to be 1998 (95% HPD 1991–2003). The NY bovine clade is further split up into two clades: (i) a clade with two isolates from northeastern New York (Figure 3.1: clade 1)

and (ii) a clade with 15 bovine associated isolates from western NY state (Figure 3.1: clade 2). The MRCA of the latter clade dates back to 2002 (95% HPD 1999–2005). Within clade 2, two well supported clusters were identified (marked 'a' and 'b' in Figure 3.1). Specifically, 'cluster a' contains six isolates that were isolated from Steuben county (NY) and the neighboring Livingston county (NY). This finding suggests a phylogeographic signal in the dataset, which should facilitate more detailed tracing of the emergence of *S*. Cerro ST367 throughout the northeastern US with a larger sampling and a population genomic analysis.

Genome sequence analysis reveals a stepwise evolution, of S. Cerro ST367 to a bovine-associated clade, characterized by deletion of selected operons and acquisition of a premature stop codon in sopA.

Loss or gain of genes within bacterial populations may indicate niche adaption of bacterial subpopulations [23]. To infer patterns of gene loss, we mapped reads of the 27 *S.* Cerro isolates against well-annotated genomes such as those of *S.* Typhimurium LT2, *S.* Typhi CT18, and *S*. Choleraesuis SC-B67. In addition we mapped the reads of *S.* Adelaide FSL A4-669 [24] against these genomes, to determine if the patterns of absence were also observed in the most recent common ancestor of this serovar and the *S.* Cerro population studied here. Reads of the 27 *S.* Cerro isolates mapped to 86, 88, and 90 % of the coding sequences in *S.* Typhi CT18, *S.* Typhimurium LT2, and *S.* Choleraesuis SC-B67, respectively. This is very similar to the percentage of genes shared (89%) between *S.* Typhimurium LT2 and *S.* Typhi CT18 [25] and falls in the higher end of the range observed by Jacobsen et al. [26] for a wide variety of *Salmonella* serovars. The genome size, and the high number of shared genes thus suggest that the lineage of *S.* Cerro studied here did not experience notable genome reduction.

Mapping of sequence reads of the isolates of the *S. Cerro* population further revealed a pattern of gene absence generally conserved within the *S.* Cerro population sampled here, suggesting that most of the genomic characteristics associated with the emergence of *S*. Cerro among bovine-associated habitats were present in the MRCA of this *S.* Cerro clade. Interestingly, some SPIs that were found here to be absent or partially absent (gene deletions) from the Cerro

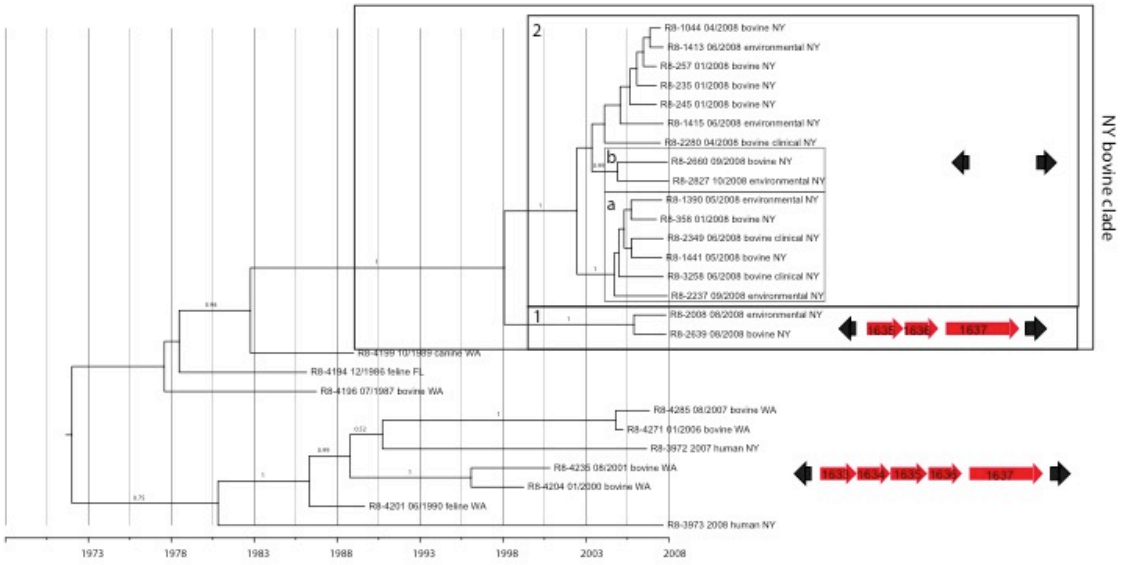

Figure 3.1 Tip-dated phylogeny of the 27 *S.* Cerro isolates sequenced in this study with stepwise deletion of a D-alanine transporter encoding gene cluster mapped onto the phylogeny. Values on the branches represent posterior probabilities. Genes adjacent to the D-alanine transporter encoding gene cluster are represented as black arrows, genes in the cluster are represented as red arrows. Numbers in the arrows refer to STM gene tags as used in the genome sequence of *S.* Typhimurium LT2. Labels on the tips indicate isolate accession numbers, isolate date (month/year) and source.

population studied (i.e., ST367), but are present in *S.* Typhimurium LT2 or *S.* Typhi CT18, have been associated with attenuation of virulence. Specifically, the genomic island at *S.* Typhi SPI-10 locus is completely absent from the *S*. Cerro ST367 isolates examined here; this SPI has been associated with virulence in mice [27]. Chaudhuri et al. [28] also showed that significant reduction of fitness of *S.* Typhimurium SL1344 is observed during intestinal colonization of cattle when genes in SPI-10 (in particular STM4489) are disrupted by transposon insertion. Genes homologous to (i) STM2230.1c to STM2240 of SPI-12, and (ii) STM3117, STM3123, and STM3119 to STM3121 of SPI-13 were also found to be absent from *S.* Cerro ST367; these SPIs have been associated with systemic infection of mice in *S*. Typhimurium [29], and replication in macrophages (SPI-13: [30]). Furthermore, disruption of STM2231 in SPI-12 and STM3123 in SPI-13 was previously shown to cause significant reduction in fitness in *S.* Typhimurium SL1344 during intestinal colonization of cattle [28]. In addition homologs of STM0293, STM0294 and STM0299 are deleted in *S.* Cerro ST367, these genes are found in SPI-16, a SPI associated with intestinal persistence in mice [31]. Disruption of STM0293 in *S.* Typhimurium has been shown to cause reduced fitness with regard to intestinal colonization of cattle [28]. Most of the SPI-related genes found to be absent in *S*. Cerro ST367 were confirmed to be present in *S.* Adelaide FSL A4-669, suggesting loss of these genes/SPIs occurred after the divergence of *S*. Adelaide from the most recent common ancestor of *S.* Cerro ST367. We found evidence for the presence of four complete TA modules (STM 2954.1N-2955.S; STM4030.S-4031; STM3777-78 and STM4449-50) within the *S.* Cerro genomes studied here. This is interesting as De la Cruz et al. [32] suggested that toxin-antitoxin (TA) modules in *Salmonella* play a role in virulence, and that the number of genomically encoded TA modules is correlated with pathogenicity of individual strains. By comparison, the number of TA modules in *S. enterica* subsp. *enterica* ranges from 5 (*S.* Paratyphi B SPB7) to 10 (*S.* Typhimurium LT2), making *S.* Cerro ST367 one of the subsp. *enterica* serotypes with the lowest number of TA modules. The number of TA modules in *S*. Cerro ST367 is similar to that observed in *Salmonella enterica* subsp. *arizonae*, a subspecies which is predominantly found in cold blooded hosts and

does generally not seem to cause illness in warm blooded hosts [33]. Complete or partial absence of some SPIs in all *S.* Cerro ST367 and the low number of TA modules in the genome, thus suggests a putative shift of *S.* Cerro in host and/or tissue tropism before the emergence of the NY bovine-associated clade.

The hypothesis that the *S.* Cerro population studied here shows unique host and/or tissue tropism characteristics is also supported by the finding that all 27 Cerro ST367 isolates sequenced here were found to carry a premature stop codon in *sopA*, causing a truncation of the gene from 782 aa (in *S*. Typhimurium LT2) to 433 aa. Previous studies have shown that SopA is involved in virulence during bovine gastrointestinal infections by *S.* Typhimurium and *S*. Dublin [34, 35], and that *sopA* mutations are implicated in reduced polymorphonuclear (PMN) cell migration [34, 36], and fluid secretion in ileal loops in calves [34]. Premature stop codons in *sopA* have been found in *S*. Typhi, *S*. Paratyphi A, and *S.* Gallinarum and it has been suggested that loss of a functional SopA has been an important factor in the virulence and adaptation of these serovars to a systemic niche in certain hosts [37, 38].

Interestingly, the one base-pair insertion responsible for the premature stop codon occurs within a ~10 bp region of *sopA* that also contains deletions in *S*. Typhi and *S*. Paratyphi A (Figure 3.2). While *S.* Typhi and *S*. Paratyphi A contain additional mutations that may have caused loss of function of SopA [38], the occurrence of deletions in the same region in *S.* Cerro *sopA* suggests this is a replication error prone region in the genome. A conserved domain search (http://www.ncbi.nlm.nih.gov/Structure/cdd/wrpsb.cgi) against the Conserved Domain Database [39] of the aa sequence of the truncated SopA in *S.* Cerro ST367 revealed the premature stop is situated in the SopA central domain [40] of the gene. Furthermore, the truncated SopA protein lacks the capsase-3 cleavage sites, which have been demonstrated to be important in induction of PMN transepithelial migration in *S.* Typhimurium [36]. Although specifically the disruption of the main functional domain in SopA and the loss of the capsase-3 cleavage sites suggest loss of function of SopA in *S*. Cerro ST367, further molecular genetic experiments have to be conducted to reveal if truncation of SopA in *S*. Cerro ST367 has lead to loss of function of this gene, and

how it affects host cell invasion (as suggested by Raffatelu et al. [41]) and other SopA associated aspects of *Salmonella* virulence.

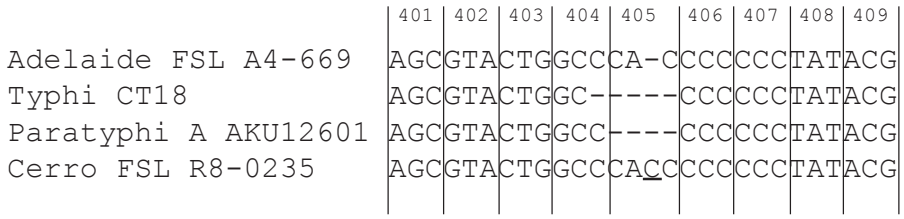

Figure 3.2 Alignment of *sopA* in *S.* Cerro and selected other *Salmonella* serovars showing premature stop codon in Cerro and *sopA* polymorphisms in other *Salmonella* strains and serovars. Numbers above the alignment indicate the amino acid residues as found in *sopA* in *S.* Typhimurium LT2. *sopA* for *S.* Adelaide FSL A4-669 is in frame, while *sopA* for *S.* Typhi CT18 and *S.* Paratyphi A AKU 12601 show a four and three bp deletion in this region in this region, respectively. *S.* Cerro has a one bp insertion (underlined), leading to a frame shift and premature stop.

Read mapping also showed one gene cluster to be stepwise deleted in the NY bovine clade (Figure 3.1). This gene cluster contains homologs of the *S.* Typhimurium LT2 genes STM1633 to STM1637. This gene cluster encodes a D-alanine transporter and has been recently shown to be required for intracellular survival in murine macrophage-like cells [42], and disruption of STM1637 has been shown to cause a significant reduction in fitness in intestinal colonization in cattle in *S.* Typhimurium [28]. This gene cluster is present in all 10 Cerro ST367 isolates that do not belong to the bovine clade. Two isolates (FSL R8-2008, FSL R8-2639) lack two genes (STM1633, STM1634) in this gene cluster. These two isolates represent a clade that split off early from the remaining NY bovine-associated-population. The remaining 15 isolates in this clade lack the entire gene cluster (Figure 3.1). The (partial) absence of the D-alanine

transporter gene cluster is currently the only genomic feature that differentiates the NY bovine clade from the remaining population (including isolates from the NY human cases).

S. Cerro displays reduced invasiveness of human epithelial cells compared to other Salmonella serovars commonly isolated from bovine sources.

The comparative genomic analyses described above suggest *S.* Cerro lacks several functional genes and genomic elements that are involved in invasion and intracellular survival. To assess if strains of *S.* Cerro ST367 population (Table 3.1) studied here are impaired in their ability to invade human intestinal epithelial cells, Caco-2 cells were infected with *S.* Typhimurium ($n = 4$), *S.* Newport ($n = 4$), *S.* Kentucky ($n = 4$), and *S.* Cerro ($n = 4$) (Appendix Table 4). Each serovar was represented by one isolate each from a bovine clinical case, a subclinically infected bovine host, an environmental sample and a human clinical case. *S.* Cerro isolates were significantly less invasive than isolates of serovars Typhimurium ($P < 0.0001$) and Newport ($P < 0.0001$), but not significantly different from *S.* Kentucky ($P = 0.0734$) (Figure 3.3).

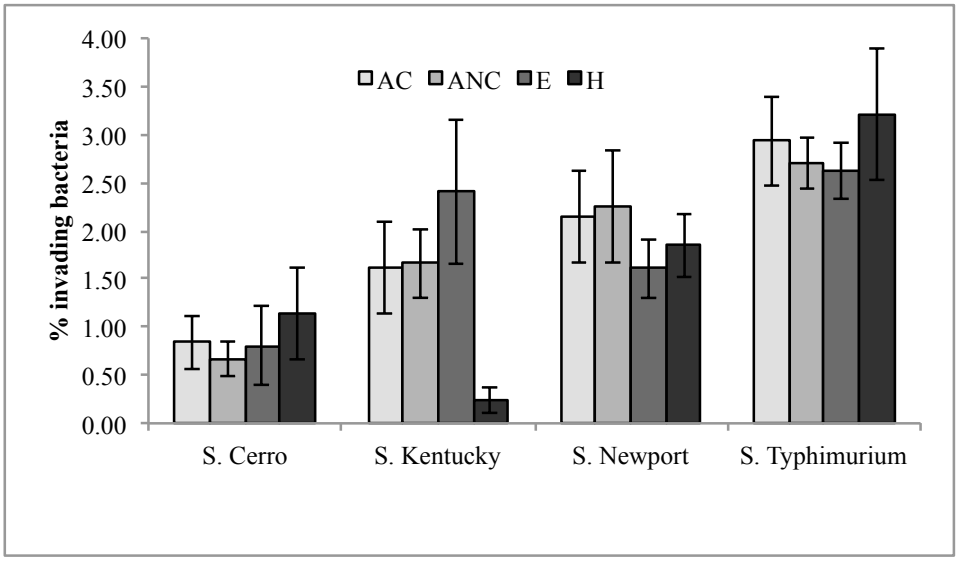

Figure 3.3 Caco-2 invasion efficiencies among *Salmonella* serovars Cerro, Kentucky, Newport, and Typhimurium. Cells were inoculated at a MOI of 10 and the invasion assays

were performed at 37°C and 5% CO_2. Invasion efficiency was calculated as [CFU recovered/CFU infected]×100. Data represent the mean of at least three biological replicates, and the error bars represent the standard deviation. The invasion efficiencies for each serovar were analyzed using one-way analysis of variance (ANOVA) and Tukey's post hoc test, after the data was log-transformed to satisfy ANOVA assumptions of normality. Isolate sources are abbreviated as AC, Animal Clinical; ANC, Animal Non-clinical; E, Environmental; H, Human.

However, the overall invasiveness of *S.* Kentucky seems to be skewed by the presence of one isolate from a human clinical case, which shows very low invasion. When this outlier is excluded from the analysis, the *S.* Cerro isolates are also significantly less invasive than *S.* Kentucky ($P = 0.004$). Thus, consistent with our genomic analyses, *S.* Cerro ST367 seems to be less invasive in human intestinal epithelial cells than the serovars examined here. Future studies on the ability of *S.* Cerro to invade bovine intestinal epithelial cells and to cause illness in cattle will be necessary though to determine whether *S.* Cerro or specific subtypes within *S.* Cerro truly show attenuated bovine virulence.

CONCLUSIONS

Comparative genomic analyses of 27 *Salmonella* Cerro isolates indicate that this serovar lacks several genes that have previously been shown to be involved in the ability of *Salmonella* serovars to cause intestinal infection. Reduced invasion of human intestinal epithelial cells, as compared to other serovars, further supports the reduced ability of this serovar to cause intestinal infection, however, further experiments are necessary to determine which genes are responsible for this phenotype. Altogether, these results suggest that the emergence of *S.* Cerro ST367 among livestock operations in the northeastern United States may not be due to increased adaptation to the bovine host, nor to increased antibiotic resistance. Instead, the frequent isolation of this serovar on cattle farms [8] may reflect that this serovar was able to dispersed rapidly as no efforts were undertaken to control its spread (possibly due to a lack of clinical

signs, which left infections undetected). Alternatively, or in addition, *S.* Cerro (or some subtypes within *S.* Cerro) may have unique phenotypic characteristics that were not discovered through the comparative genomic analyses conducted here, but that facilitate environmental survival or dispersal.

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CHAPTER 4

IDENTIFICATION OF FUNCTIONAL CYTOLETHAL DISTENDING TOXIN B (TYPHOID TOXIN) IN NONTYPHOIDAL *SALMONELLA* SEROVARS*

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ABSTRACT

Salmonella strains encode an extensive repertoire of virulence genes and disease severity varies between serovars. Here we show that a functional typhoid CdtB-islet, closely related to the *S.* Typhi CdtB-islet, is present in a monophyletic clade of nontyphoidal *Salmonella* serovars. At 72 h after infection, Henle-407 cells infected with nontyphoidal *Salmonella* strains that encode CdtB displayed arrest in G_2/M, while cells infected with the isogenic CdtB null mutants did not display a G_2/M phase arrest. These results indicate that a number of nontyphoidal *Salmonella* serovars encode a CdtB toxin that leads to cell cycle arrest, suggesting a possible role of CdtB in host-pathogen interaction.

INTRODUCTION

Cytolethal distending toxin B (CdtB) is a recently recognized virulence factor that is found in *Salmonella enterica* serovar Typhi [1, 2] as well as a number of other Gram negative bacterial pathogens [3]. Host cells intoxicated with CdtB undergo irreversible cell cycle arrest in response to DNA damage which, if severe, leads to cell death by apoptosis [3]. Recent work by our group identified an islet encoding the cytolethal distending toxin B (CdtB-islet) in the genomes of 50 nontyphoidal *Salmonella*; this islet was found in 37/38 isolates that were classified into a clade designated "Clade B", in isolates representing serovars Typhi, Paratyphi A, 8/115 nontyphoidal clade A isolates, and in five isolates that did not clearly group into clades A or B [4]. The CdtB-islet in *Salmonella* Typhi includes five genes (i.e., *pltA*, *pltB*, *ttsA*, *sty1887*, in addition to *cdtB*). *pltA* and *pltB* encode homologs to pertussis toxin components [5] responsible for ADP-ribosylation of a host protein [5] and export of CdtB from the *Salmonella* containing vacuole as well as from the host cell; *ttsA* encodes a phage-origin muramidase necessary for the secretion of the PltA/CdtB/PltB toxin [6]. *sty1887* encodes a putative homolog of a phage tail protein; deletion of this gene in *Salmonella* Typhi did not have an effect on typhoid toxin secretion [6].

Due to the well-documented role of CdtB in host-pathogen interactions in *Salmonella* Typhi and other bacterial pathogens, we hypothesized that CdtB may also play a role in the

interaction of nontyphoidal *Salmonella* serovars and host cells. We therefore, used a comparative genomics approach to characterize sequence conservation of *plt*A, *plt*B, and *cdt*B among *S.* Typhi and nontyphoidal *Salmonella* serovars. We subsequently created CdtB null mutants in three nontyphoidal *Salmonella* strains in order to assess the function of CdtB in these strains, using Henle-407 cells infection and host cell cycle analysis to specifically test the hypothesis that CdtB in these strains causes cell cycle arrest, similar to the CdtB function that has been reported for *S.* Typhi.

MATERIALS AND METHODS

PltA, PltB, and CdtB phylogeny and amino acid sequence analysis

PltA, PltB and CdtB amino acid sequences for *S.* Typhi CT18 were obtained from GenBank. In addition to a protein homology BLAST search, *Salmonella* genomes (both finished and draft) were queried for homologs of coding nucleotide sequences of S. Typhi CT18 PltA, PltB and CdtB using blastn. The nucleotide sequences obtained from the blast search were translated into amino acid sequences and aligned using MAFFT [7], employing the 'auto' alignment strategy. Maximum likelihood phylogenies were created with PhyML (version 20130708), using the WAG model of amino acid substitution and a gamma distribution of variable sites. To assess the robustness of the inferred phylogeny 250 bootstrap replicates were performed for each analysis.

Bacterial cultures

Three *Salmonella* isolates representing serovars Javiana (isolate FSL S5-0395), Schwarzengrund (isolate FSL R6-879), and Montevideo (isolate FSL R8-4841) were obtained from the New York State Department of Health (NYSDOH); all isolates were from humans with clinical symptoms consistent with salmonellosis. Presence of *plt*A, *plt*B, and *cdt*B in these strains was confirmed using TaqMan® assays (Life Technologies, Foster City, CA) as previously described [4] to determine if strains were suitable for constructing isogenic mutant for the CdtB.

Isogenic Δ*cdtB* mutants were constructed using the Lambda Red system as previously described [8] and *cdtB* deletions were confirmed by PCR and sequencing of the deletion allele.

Epithelial cell infection

Bacterial cultures for epithelial cell infection were prepared as previously described [2], with some modifications; all *Salmonella* growth steps were performed in LB broth (with 0.3 M NaCl) at 37ºC, without shaking. Briefly, overnight *Salmonella* cultures were diluted 1:100 in fresh LB (0.3 M NaCl) broth and incubated at 37ºC until they reached OD_{600} of 0.4. Then, these cultures were diluted 1:100 into Nephelo culture flasks with 50 mL LB 0.3 M NaCl broth, and incubated at 37ºC until the cultures reached an OD_{600} of 0.4, followed by incubation for an additional 3 h (to yield approx. $1x10^9$CFU/mL).

Henle-407 cells (ATCC CCL-6) were grown at 37°C with 5.0–5.5% CO_2 in Dulbecco's Modified Eagle Medium (DMEM) (Corning, Manassas, VA) with 10% FBS (Atlanta Biologicals, Lawrenceville, GA). For infection experiments, cells were seeded in 6-well plates and incubated for 22–23 h before inoculation. Media in the 6-well plate was replaced with fresh DMEM 10% FBS 30 minutes before the wells were inoculated with *Salmonella*. Henle-407 cells were inoculated at an MOI of 50. After incubation at 37°C (5.0–5.5% CO_2) for 1 h, cells were washed with pre-warmed phosphate buffered saline (PBS), fresh DMEM 10% FBS with gentamicin (100μg/mL) was added to each well, and the 6-well plates were incubated for 1 h. After 1 h incubation, cells were washed 3 times with pre-warmed PBS and fresh DMEM 10% FBS with gentamicin (10μg/mL) was added to each well. Cells were incubated for a total of 72 h post infection. Finally, the cells were prepared for Fluorescence Activated Cell Sorting (FACS) analysis, performed as detailed below. These assays were performed in duplicate in two separate biological replicates between passages 12–50.

Cell cycle analysis

Cell cycle analysis of infected cells as well as uninfected control cells was performed as described previously [9] with some modifications. Briefly, Henle-407 cells were washed, trypsinized, and centrifuged at 1,500 rpm for 5 min at room temperature. The supernatant was

then removed and cold 70% ethanol was added (while vortexing at slow speed), to fix the cells. After the fixed cells were kept at –20°C for at least 1 h, PBS containing 0.1% Tween 20 (Sigma-Aldrich, St. Louis, MO) and 1% BSA (Fisher Scientific, Fair Lawn, NJ) (PBST) was added to increase cell membrane permeability and to rehydrate the cells; cells were then incubated for 10 min at room temperature. Cells were then centrifuged at 1,500 rpm for 5 min, followed by removal of supernatant. After one more PBST wash, cells were re-suspended in a Propidium Iodide (PI, Sigma-Aldrich, St. Louis, MO) staining solution (40µg of PI/mL, 100µg of RNase A/mL [Sigma-Aldrich]) and incubated at room temperature in the dark for 10 min. Subsequent DNA content analysis of approximately 3.0 x10^4 cells was performed by FACS using a LSRII Flow Cytometer (BD-Biosciences, San Jose, CA) in the Biomedical Sciences Flow Cytometry Core Laboratory at Cornell University. The proportions of Henle-407 cells in G_1, S, and G_2 phase of the cell cycle were calculated after quantifying the mean percentages of the cells detected in manually adjusted gates around 2N, 3N, and 4N DNA content.

Statistical Analysis

One-way ANalysis Of VAriance (ANOVA) was used to determine whether the strain used for infection (parent strain versus Cdt isogenic mutant) had a significant ($P < 0.05$) effect on the percentage of Henle-407 cells identified to be in a given cell cycle stage (G_1, S, or G_2/M).

RESULTS

A maximum likelihood based phylogeny of CdtB amino acid sequences showed that homologs of this gene are widely distributed among Gram-negative bacteria. In addition to *S.* Typhi and *S.* Paratyphi A, homologs of the typhoid holotoxin genes were confirmed in 11 serovars classified into *S. enterica* subsp. *enterica* clade B (13 isolates, see Figure 4.1), *S. enterica* subsp. *enterica* serovar Inverness (FSL R8-3668), *S. enterica* subsp. *arizonae* (RSK2980), and *S. bongori* (NCTC 12419). All genomes that contained a gene encoding CdtB also included genes encoding two paralogs of PltA and PltB in the CdtB-islet; in *S.* Typhi these proteins are part of the typhoid holotoxin complex [10]. We also identified two other *pltA* and

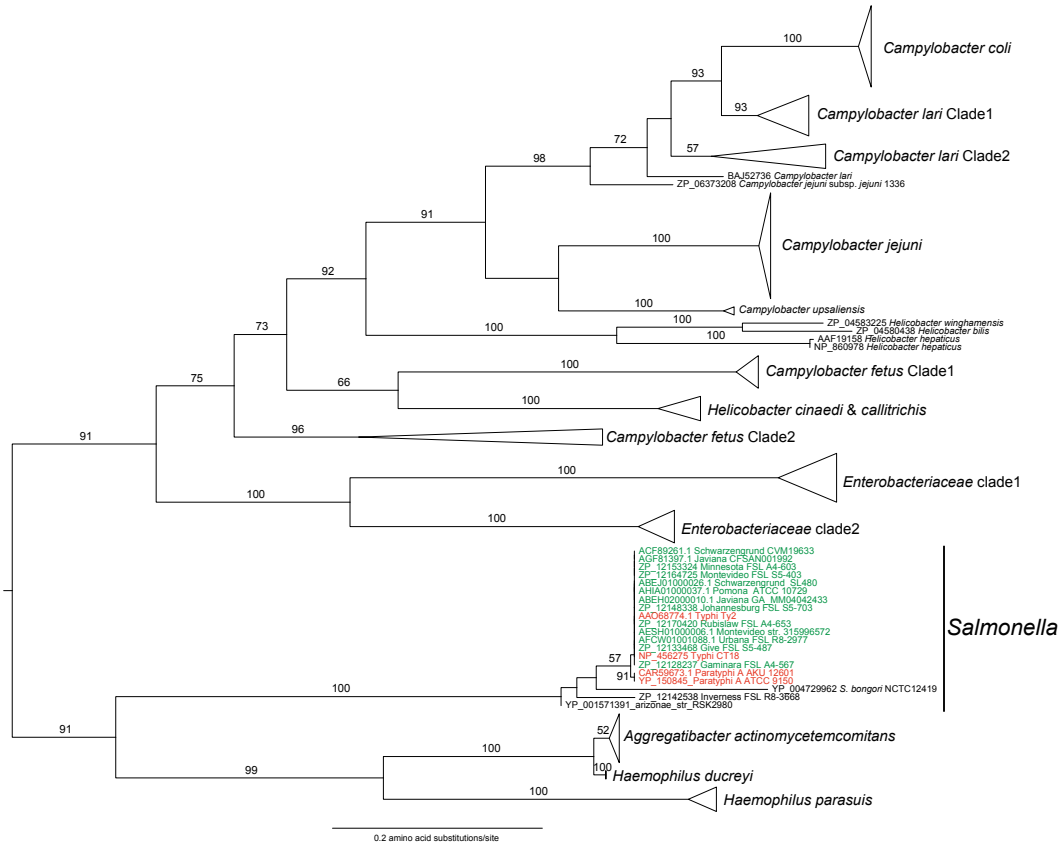

Figure 4.1 Amino acid based maximum likelihood phylogeny of CdtB. Nontyphoidal *Salmonella enterica* subsp. *enterica* serovars are colored green, while *S.* Typhi accessions are colored red. Values on or next to the branches are bootstrap values based on 250 bootstrap replicates.

pltB paralogs, which have previously been annotated as the *artA/ artB* operon in *S.* Typhimurium DT104 [11] and encode an AB5 toxin [12], in a genomic region that is not in proximity to the CdtB-islet (Appendix Figure 2 and Appendix Figure 3). *S.* Typhi CdtB, PltA, and PltB show high levels of homology with the corresponding proteins in nontyphoidal *S. enterica* subsp. *enterica* isolates (with the exception of *S.* Inverness) with 99.3 – 100% aa sequence identity for CdtB, 98.3 – 100 % aa sequence identity for PltA, and 94.9 – 100% aa sequence identity for PltB. Moreover, functionally critical residues (i.e., PltA Cys 214, CdtB Cys 269) within the typhoid holotoxin proteins, as determined by Song et al. [10], were found to be conserved in all 13 nontyphoidal *Salmonella* genomes examined here (see Appendix Table 5 for genomes).

As the sequence conservation for CdtB, PltA, and PltB along and with the conservation of functionally critical aa residues suggests that a functional typhoid holotoxin is produced by nontyphoidal salmonellae, we constructed isogenic mutants with deletions of *cdtB* in three non-typhoidal *Salmonella enterica* strains, classified in Clade B, for phenotypic characterization. When parent strains (representing serovars Javiana, Schwarzengrund, and Montevideo) and corresponding isogenic mutants were used to infect Henle-407 cells, a clear difference in the cell cycle status of the infected cells could be observed at 72 h post inoculation. The majority of uninfected control cells (60.0%) as well as the majority of cells infected with the three *Salmonella* CdtB null mutants (61.2% average across all three strains) were in G_1 phase. By comparison, a significantly smaller percentage of the Henle-407 cells infected with the parent strains were G_1 phase (18.6% average across all three parent strains; ANOVA $P < 0.0001$; Figure 4.2). On the other hand, the majority of the Henle-407 cells infected with *Salmonella* parent strains were in G_2/M phase (60.3% average across all three parent strains); the proportion of host cell in G_2/M phase was significantly higher in the cells infected with the parent strains as compared to the cell infected with the CdtB mutant strains ($P < 0.0001$, ANOVA; see Figure 4.2). These findings suggest presence of a functional CdtB that causes G_2/M phase cell arrest in cells infected with the three parent strains evaluated here.

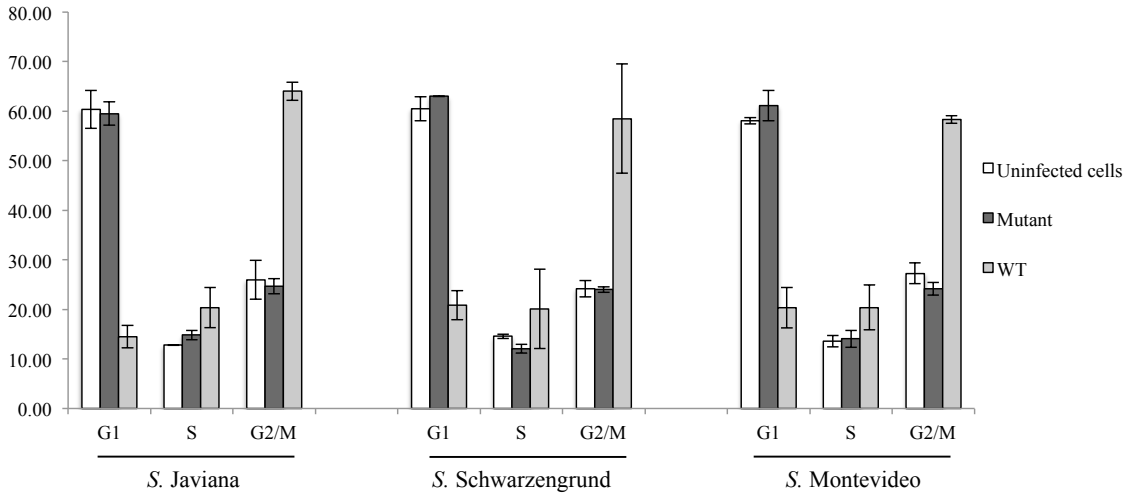

Figure 4.2 Cell cycle phase of Henle-407 cells infected with nontyphoidal *Salmonella* wild type (WT) strains and their isogenic mutants for the CdtB. Toxicity of the cells infected with *Salmonella* WT strains and their isogenic mutants were analyzed by FACS at 72 h of infection. Data shown represent the averages of two independent biological replicates with two technical replicates (two infected wells) preformed for each biological replicate. Cells infected with *Salmonella* parent strains (i.e., *S.* Montevideo, *S.* Schwarzengrund, and *S.* Javiana) showed a significantly higher proportion of cells in G_2/M phase as when compared to the isogenic D*cdtB* mutants ($P < 0.0001$ for the factor D*cdtB* deletion; ANOVA). Conversely, cells infected with the isogenic D*cdtB* mutants showed a significantly higher proportion of cells in G_1 phase ($P < 0.0001$; ANOVA) as compared to the parent strain.

DISCUSSION

A comparative genomic study by den Bakker et al. [4] previously revealed the presence of the cytolethal distending toxin B encoding islet (CdtB-islet) within the genomes of 54 non-typhoidal *Salmonella* isolates, of which the majority (70%) belonged to a restricted subpopulation of *S. enterica* subsp. *enterica* (Clade B). In addition, Desai et al. [13] also reported the presence of *cdtB* (referred to as Typhoid toxin in that study) in the genomes of 2 *S. enterica* subsp. *diarizoniae* and 2 *S. enterica* subsp. *arizoniae* strains. Consistent with previous analyses [14, 15], our comparative genomic analyses also showed that homologs of *Salmonella* Typhi *cdtB* are widely distributed among Gram-negative bacterial pathogens. Contributions of CdtB to virulence or host – pathogen interactions also have been reported in a number of bacterial pathogens, including reduced cytotoxicity in CdtB null mutants (e.g., *H. ducreyi*, *H. hepaticus*, *C. jejuni*) and CdtB mediated G_2/M cell arrest (e.g., *E. coli*, *A. actinomycetemcomitans*, *H. ducreyi*) (3). Importantly, our sequence analysis showed that genes found in the CdtB islet of *S.* Typhi and nontyphoidal serovars are highly conserved and share a common phylogenetic ancestor, providing initial evidence suggesting presence of a functional CdtB in a subset of nontyphoidal *Salmonella*.

Phenotypic characterization of CdtB null mutants in three serovar backgrounds showed that presence of the CdtB is critical to cause G_2/M phase cell arrest in infected Henle-407 cells. In contrast, previous studies only evaluated the contributions of CdtB to host cell – pathogen interactions in *S.* Typhi [5, 6, 10]; these studies found that *S.* Typhi CdtB also is responsible for G_2/M phase cell arrest in infected cells. Our data reported here suggest, for the first time, a role for CdtB in a number of nontyphoidal *Salmonella* strains. Our findings also suggest the possibility that nontyphoidal *Salmonella* strains may represent distinct subgroups that differ in their interactions with host cells, with CdtB-islet positive strains possible causing cell arrest in cells in the infected target tissues, which may increase the risk of long-term sequelae that could be associated with G_2/M cell cycle arrest. For example, Lara-Tejero [9] has suggested that

exposure to CdtB and the associated DNA damage caused by this toxin in individuals infected with *C. jejuni* may be a predisposing factor to intestinal cancer.

ACKNOWLEDGMENTS

We would like to acknowledge Dr. Rasika Jinadasa and Lavanya Gowri Sayam for the training provided on sample processing for FACS analysis and FACS analysis, respectively. This work was supported in part by a USDA NIFA Special Research Grant to MW (2010-34459-20756).

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CHAPTER 5

CONCLUSION

Salmonella is a major public health concern. The hosts and reservoirs of *Salmonella* are extremely diverse and its ability to survive in the environment makes this pathogen difficult to control or eradicate. Our studies shed light on nontyphoidal *Salmonella* serovars and on the virulence factors that play a role in the pathogenicity and host specificity of this pathogen.

Our first objective was to characterize the diversity of *Salmonella* isolated from subclinical dairy cattle and dairy farm environments. Our data indicate that subclinical livestock and farm environments may represent a potentially important reservoir of *Salmonella* serovars and subtypes associated with human infections, including MDR *Salmonella*. However, a number of *Salmonella* serovars commonly found among subclinically infected cattle are rare among human clinical cases, for example, *Salmonella* Cerro. Serovars Cerro and Kentucky accounted for more than 50% of the serovars isolated during our study. Interestingly, in recent years, *Salmonella* Cerro has been described as the most commonly isolated serovar from healthy dairy cattle in the U.S. Animals shedding *Salmonella* cannot be recognized through clinical signs, which reduces the likelihood of adequate biosecurity efforts and quarantine efforts for these non-clinical shedders. Because subclinical shedders are able to spread *Salmonella* into the environment, they represent a risk of within-herd transmission but more importantly a public health concern. Future work is necessary to improve the understanding of the dynamics of infection in order to control or mitigate *Salmonella* in livestock and livestock-associated environments.

In our second study, we decided to investigate the genomic features that could facilitate the pervasiveness of *Salmonella* Cerro in dairy cattle and the farm environments, but that allow this serovar to be so rare among humans. We hypothesized that features in the *Salmonella* Cerro

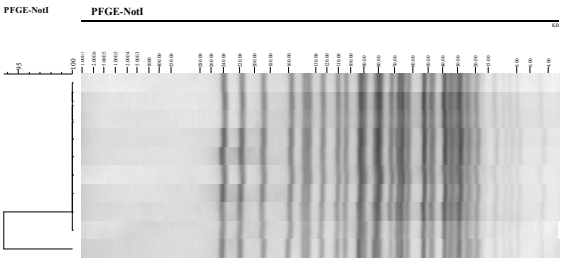
genome were likely to be the cause of its unique epidemiological characteristics. For this objective we used comparative and population genomic tools to study 27 *Salmonella* Cerro isolates from different geographic locations in the U.S. (i.e., New York, Washington, Florida). Our data indicate that all 27 sequenced isolates represented sequence type ST367, and that there is a well-supported clade (NY bovine clade) within the *S.* Cerro population represented here. The most recent common ancestor of the NY bovine clade dates back to 1998, according to the molecular clock analysis performed, and these results support the hypothesis of a recent emergence of this clone. Our study revealed that the *Salmonella* Cerro isolates lack several genes that have been previously described as necessary for gastrointestinal infections. In addition, we have demonstrated that this serovar has a reduced ability to invade human epithelial cells when compared to other serovars (i.e., Typhimurium, Newport, and Kentucky). This further supports the hypothesis of its reduced ability to cause intestinal infection predicted from the whole genome sequencing data. Future studies on the ability of *Salmonella* Cerro to invade bovine intestinal epithelial cells and to cause illness in cattle will be necessary though to determine whether *Salmonella* Cerro or specific subtypes within *Salmonella* Cerro truly show attenuated bovine virulence. Next generation sequencing technologies are readily accessible and have proven to be great tools for the study and understanding of emerging pathogens and strains responsible for harmful outbreaks.

Our third objective was built-on previous genomic studies by our research group, which revealed that the islet encoding the cytolethal distending toxin B (CdtB-islet) was found in the genomes of 50 nontyphoidal *Salmonella*. We hypothesized that the cytolethal distending toxin B (CdtB) encoded in this pathogenicity islet was functional and may play a role in the interaction of nontyphoidal *Salmonella* serovars and host cells. A characterization of sequence conservation

of *cdt*B, *plt*A, and *plt*B, among *Salmonella* Typhi and nontyphoidal *Salmonella* serovars, showed that these sequences as well as their functionally critical amino acid residues are conserved, which suggested that a functional typhoid holotoxin is produced by nontyphoidal *Salmonella*. Henle-407 cells infected with nontyphoidal *Salmonella* strains that encode CdtB displayed arrest in G_2/M, while cells infected with the isogenic CdtB null mutants did not display arrest in G_2/M. Our findings suggest, for the first time, a role for CdtB in serovars other than *Salmonella* Typhi. Host cells exposed to CdtB-islet positive strains may be more severely affected than host cells infected with CdtB-islet negative strains, because of the risk of long-term sequelae that could be associated with cell cycle arrest.

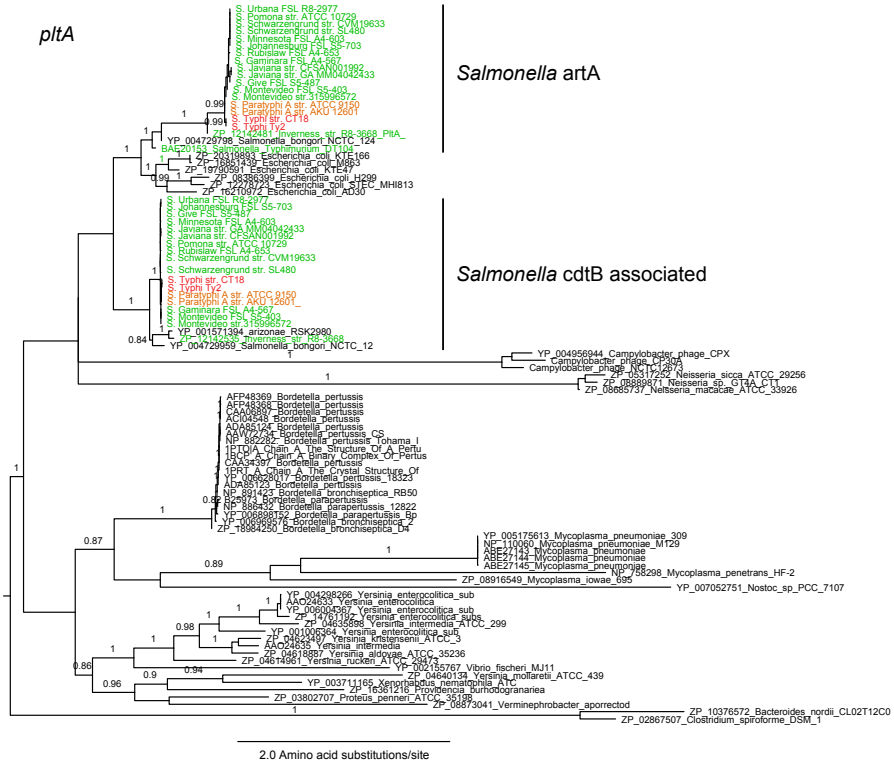
This work adds to the growing body of scientific research on *Salmonella* virulence and host interaction. High-throughput next generation sequencing in conjunction with comparative and population genomic analyses are powerful approaches, which will allow future research efforts to investigate multiple genomic features simultaneously in larger sets of genomes. These approaches may provide a more comprehensive understanding of *Salmonella* serovars in the near future, in contrast to other approaches (e.g., serotyping, MLST, etc.) that are only focused on specific phenotypic and genotypic characteristics. Novel functions and mechanisms of *Salmonella* virulence and genomic factors uncovered by bioinformatics approaches may need to be confirmed using animal models in order to expand the body of knowledge on the virulence, pathogenicity, and host specificity of *Salmonella* spp. These studies will shed light on different aspects of *Salmonella* and will facilitate tracking the source of outbreaks, and the development of better diagnostic tools in order to mitigate and control this pathogen.

APPENDIX

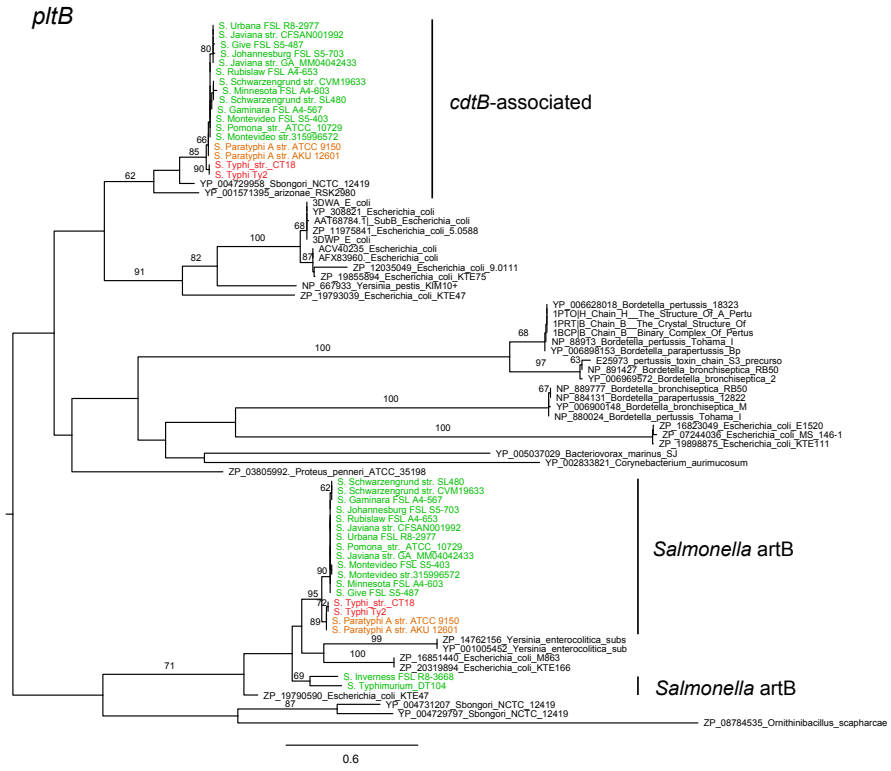


FSL No.	Source	Farm ID	*Not*I PFGE Type No.	*Xba*I PFGE Type No.
FSL R8-072	Bovine Non-clinical	26	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-1397	Environmental	49	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-1415	Environmental	60	NYCUJAAN11.0002	NYCUJAAX01.0973
FSL R8-2034	Environmental	47	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-2036	Environmental	65	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-245	Bovine Non-clinical	30	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-2660	Bovine Non-clinical	65	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-2822	Environmental	65	NYCUJAAN11.0002	NYCUJAAX01.0213
FSL R8-2856	Environmental	54	NYCUJAAN11.0002	NYCUJAAX01.0840
FSL R8-2639	Bovine Non-clinical	52	NYCUJAAN11.0003	NYCUJAAX01.0843

Appendix Figure 1 *Not*I-PFGE types for 10 *Salmonella* Cerro isolates represent 4 different *Xba*I-PFGE types. These isolates were differentiated into 2 *Not*I-PFGE types.



Appendix Figure 2 PltA amino acid phylogeny. Amino acid based maximum likelihood phylogeny of *pltA*. Nontyphoidal *Salmonella enterica* subsp. *enterica* serovars are colored green, *S.* Paratyphi A accessions are colored orange, and *S.* Typhi accessions are colored red. Values on or next to the branches are bootstrap values based on 250 bootstrap replicates.



Appendix Figure 3 PltB amino acid phylogeny. Amino acid based maximum likelihood phylogeny of *pltB*. Nontyphoidal *Salmonella enterica* subsp. *enterica* serovars are colored green, *S.* Paratyphi A accessions are colored orange, and *S.* Typhi accessions are colored red. Values on or next to the branches are bootstrap values based on 250 bootstrap replicates.

Supplemental Table 1. List of the 1,349 *Salmonella* isolates obtained from dairy cattle and dairy farm environments from 46 NY state farms.

Entry	Unique ID	FSL Num	Farm ID	Source	Serovar[a]	Sampling Date	Antimicrobial susceptibility pattens	Subjected to PFGE typing[b]	PFGE type number
50	LR-1032-01	FSL R6-938	1	Environmental	Meleagridis	10/2/07	CEF1	Y	CU-175
63	LR-1035-01	FSL R6-951	1	Bov non-clin	Meleagridis	10/2/07	PAN0	N	
64	LR-1035-02	FSL R6-952	1	Bov non-clin	Meleagridis	10/2/07	PAN0	Y	CU-175
65	LR-1036-01	FSL R6-953	1	Bov non-clin	Meleagridis	10/2/07	CEF1	Y	CU-175
66	LR-1037-01	FSL R6-954	1	Bov non-clin	Typhimurium	10/2/07	PAN0	Y	CU-655
67	LR-1038-01	FSL R6-955	1	Bov non-clin	Muenster	10/2/07	PAN0	Y	CU-1030
29	LR-1022-01	FSL R6-802	1	Bov non-clin	Meleagridis	10/30/07	AMP1 CEF1 CRO1	N	
30	LR-1023-01	FSL R6-803	1	Bov non-clin	Meleagridis	10/30/07	PAN0	N	
31	LR-1023-02	FSL R6-804	1	Bov non-clin	Meleagridis	10/30/07	PAN0	N	
32	LR-1022-02	FSL R6-805	1	Bov non-clin	Meleagridis	10/30/07	AMP1 CEF1 CRO1	Y	CU-175
33	LR-1023-03	FSL R6-806	1	Bov non-clin	Meleagridis	10/30/07	PAN0	N	
34	LR-1022-03	FSL R6-807	1	Bov non-clin	Meleagridis	10/30/07	AMP1 CEF1 CRO1	N	
35	LR-1023-04	FSL R6-808	1	Bov non-clin	Meleagridis	10/30/07	PAN0	N	
36	LR-1023-05	FSL R6-809	1	Bov non-clin	Meleagridis	10/30/07	PAN0	Y	CU-175
37	LR-1024-01	FSL R6-810	1	Bov non-clin	Meleagridis	10/30/07	CEF2	Y	CU-175
197	LR-1086-01	FSL R8-192	1	Bov non-clin	Typhimurium Copenhagen	12/11/07	PAN0	N	
198	LR-1086-02	FSL R8-193	1	Bov non-clin	Typhimurium Copenhagen	12/11/07	PAN0	Y	CU-71
199	LR-1086-03	FSL R8-194	1	Bov non-clin	Typhimurium Copenhagen	12/11/07	PAN0	N	
200	LR-1087-01	FSL R8-195	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
201	LR-1087-02	FSL R8-196	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
202	LR-1087-03	FSL R8-197	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
203	LR-1087-04	FSL R8-198	1	Bov non-clin	Meleagridis	12/11/07	PAN0	Y	CU-175
204	LR-1087-05	FSL R8-199	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
205	LR-1087-06	FSL R8-200	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
206	LR-1087-07	FSL R8-201	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
207	LR-1087-08	FSL R8-202	1	Bov non-clin	Meleagridis	12/11/07	PAN0	N	
169	LR-1071-01	FSL R8-164	1	Environmental	Meleagridis	1/10/08	PAN0	N	
170	LR-1071-02	FSL R8-165	1	Environmental	Meleagridis	1/10/08	PAN0	Y	CU-175
171	LR-1071-03	FSL R8-166	1	Environmental	Meleagridis	1/10/08	PAN0	N	
172	LR-1071-04	FSL R8-167	1	Environmental	Meleagridis	1/10/08	PAN0	N	
351	LR-1140-01	FSL R8-901	1	Environmental	Meleagridis	4/4/08	PAN0	Y	CU-175
509	LR-1169-01	FSL R8-1381	1	Environmental	Meleagridis	5/8/08	PAN0	Y	CU-175
510	LR-1169-02	FSL R8-1383	1	Environmental	Meleagridis	5/8/08	PAN0	N	
511	LR-1169-03	FSL R8-1384	1	Environmental	Meleagridis	5/8/08	PAN0	N	
533	LR-1182-01	FSL R8-1406	1	Environmental	Meleagridis	6/13/08	PAN0	Y	CU-175
625	LR-1205-01	FSL R8-1633	1	Environmental	Meleagridis	7/23/08	PAN0	Y	CU-175
626	LR-1205-02	FSL R8-1634	1	Environmental	Meleagridis	7/23/08	PAN0	N	
627	LR-1205-04	FSL R8-1636	1	Environmental	Meleagridis	7/23/08	PAN0	N	
857	LR-1250-01	FSL R8-2233	1	Environmental	Meleagridis (Multiple serovars)	9/10/08	PAN0	E	
858	LR-1250-02	FSL R8-2234	1	Environmental	Meleagridis	9/10/08	PAN0	Y	CU-175
859	LR-1250-03	FSL R8-2235	1	Environmental	Meleagridis	9/10/08	PAN0	N	
1079	LR-1297-01	FSL R8-2829	1	Environmental	Meleagridis* (Untypeable)	10/22/08	PAN0	Y	CU-175
1080	LR-1298-01	FSL R8-2830	1	Environmental	3,10:-:1,w	10/22/08	PAN0	Y	CU-175
106	LR-1055-01	FSL R8-345	1	Environmental	Meleagridis	Not given	PAN0	Y	CU-175
107	LR-1055-02	FSL R8-346	1	Environmental	Meleagridis (Kentucky)	Not given	PAN0	E	
289	LR-1115-01	FSL R8-822	3	Bov non-clin	Muenster	2/7/08	PAN0	Y	CU-836
617	LR-1199-01	FSL R8-1625	4	Environmental	Anatum	7/15/08	AMC1 AMP1 FOX1 CEF1 CRO2	Y	CU-1026
955	LR-1270-01	FSL R8-2634	4	Bov non-clin	Anatum	8/18/08	AMC1 AMP1 FOX1 CEF1 CRO2 TEL1	Y	CU-1026
48	LR-1031-01	FSL R6-936	10	Environmental	Agona	11/27/07	AMC1 AMP1 FOX1 CEF1 CHL1 KAN1 STR1 SUL1 TEL1 SXT1	N	
49	LR-1031-02	FSL R6-937	10	Environmental	Agona	11/27/07	AMC1 AMP1 FOX1 CEF1 CHL1 KAN1 STR1 SUL1 TEL1 SXT1	Y	CU-807
519	LR-1173-01	FSL R8-1392	10	Environmental	Infantis	5/28/08	PAN0	Y	CU-261
543	LR-1188-01	FSL R8-1416	10	Environmental	Oranienburg	6/30/08	AMC1 AMP1 CHL2 GEN2 TEL1	Y	CU-844
544	LR-1189-01	FSL R8-1417	10	Environmental	Oranienburg (Cerro)	6/30/08	PAN0	Y	CU-844
770	LR-1229-01	FSL R8-2013	10	Environmental	Oranienburg	9/1/08	PAN0	Y	CU-844
771	LR-1230-01	FSL R8-2014	10	Environmental	Mbandaka	9/1/08	PAN0	Y	CU-757
28	LR-1021-01	FSL R6-801	11	Environmental	Meleagridis	10/31/07	PAN0	Y	CU-175

Appendix Table 1

618	LR-1200-01	FSL R8-1626	11	Environmental	Meleagridis	7/18/08	PAN0	Y	CU-175
767	LR-1227-01	FSL R8-2010	11	Environmental	Meleagridis	8/25/08	PAN0	Y	CU-175
867	LR-1256-01	FSL R8-2243	11	Environmental	Meleagridis	9/28/08	PAN0	N	
868	LR-1256-02	FSL R8-2244	11	Environmental	Meleagridis	9/28/08	PAN0	Y	CU-1028
1084	LR-1300-01	FSL R8-2834	11	Environmental	3,10:e,h:-	10/30/08	PAN0	Y	CU-175
1085	LR-1301-01	FSL R8-2835	11	Environmental	Meleagridis	10/30/08	PAN0	N	
1086	LR-1301-02	FSL R8-2836	11	Environmental	Meleagridis	10/30/08	PAN0	Y	CU-175
25	LR-1018-01	FSL R6-798	14	Environmental	Kentucky	10/11/07	PAN0	Y	CU-96
26	LR-1019-01	FSL R6-799	14	Environmental	Newport* (Kentucky)	10/11/07	PAN0	Y	CU-121
54	LR-1034-01	FSL R6-942	14	Bov non-clin	Kentucky	11/6/07	PAN0	Y	CU-96
55	LR-1034-02	FSL R6-943	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
56	LR-1034-03	FSL R6-944	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
57	LR-1034-04	FSL R6-945	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
58	LR-1034-05	FSL R6-946	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
59	LR-1034-06	FSL R6-947	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
60	LR-1034-07	FSL R6-948	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
61	LR-1034-08	FSL R6-949	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
62	LR-1034-09	FSL R6-950	14	Bov non-clin	Kentucky	11/6/07	PAN0	N	
161	LR-1069-01	FSL R8-083	14	Bov non-clin	Kentucky	12/13/07	PAN0	Y	CU-96
162	LR-1069-02	FSL R8-084	14	Bov non-clin	Kentucky	12/13/07	PAN0	N	
208	LR-1069-03	FSL R8-203	14	Bov non-clin	Kentucky	12/13/07	PAN0	N	
209	LR-1069-04	FSL R8-204	14	Bov non-clin	Kentucky	12/13/07	PAN0	N	
210	LR-1069-05	FSL R8-205	14	Bov non-clin	Kentucky	12/13/07	PAN0	N	
104	LR-1054-01	FSL R8-343	14	Environmental	Kentucky	2/14/08	PAN0	N	
105	LR-1054-02	FSL R8-344	14	Environmental	Kentucky	2/14/08	PAN0	Y	CU-96
306	LR-1123-01	FSL R8-839	14	Bov non-clin	Kentucky	2/14/08	PAN0	N	
307	LR-1123-02	FSL R8-840	14	Bov non-clin	Kentucky	2/14/08	PAN0	Y	CU-96
772	LR-1231-01	FSL R8-2015	14	Environmental	Kentucky	8/30/08	PAN0	N	
773	LR-1231-02	FSL R8-2016	14	Environmental	Kentucky	8/30/08	PAN0	Y	CU-96
27	LR-1020-01	FSL R6-800	15	Environmental	Kentucky	10/11/07	PAN0	Y	CU-96
51	LR-1033-01	FSL R6-939	15	Bov non-clin	Kentucky	11/5/07	PAN0	N	
52	LR-1033-02	FSL R6-940	15	Bov non-clin	Kentucky	11/5/07	PAN0	N	
53	LR-1033-03	FSL R6-941	15	Bov non-clin	Kentucky	11/5/07	PAN0	Y	CU-96
163	LR-1070-01	FSL R8-085	15	Bov non-clin	Kentucky	12/14/07	PAN0	N	
164	LR-1070-02	FSL R8-086	15	Bov non-clin	Kentucky	12/14/07	PAN0	N	
165	LR-1070-03	FSL R8-087	15	Bov non-clin	Kentucky	12/14/07	PAN0	Y	CU-96
166	LR-1070-04	FSL R8-088	15	Bov non-clin	Kentucky	12/14/07	PAN0	N	
167	LR-1070-05	FSL R8-089	15	Bov non-clin	Kentucky	12/14/07	PAN0	N	
168	LR-1070-06	FSL R8-090	15	Bov non-clin	Kentucky	12/14/07	PAN0	N	
100	LR-1051-01	FSL R8-339	15	Environmental	Kentucky	2/12/08	PAN0	Y	CU-96
101	LR-1051-02	FSL R8-340	15	Environmental	Kentucky	2/12/08	PAN0	N	
299	LR-1119-01	FSL R8-832	15	Bov non-clin	Kentucky	2/12/08	PAN0	N	
300	LR-1119-02	FSL R8-833	15	Bov non-clin	Kentucky	2/12/08	PAN0	Y	CU-96
301	LR-1119-03	FSL R8-834	15	Bov non-clin	Kentucky	2/12/08	PAN0	N	
302	LR-1120-01	FSL R8-835	15	Bov non-clin	Dublin* (Untypeable Rough O:NonMotile)	2/12/08	AMP1 CHL1 KAN1 STR1 SUL1 TEL1 SXT1	Y	CU-585
1096	LR-1305-01	FSL R8-2846	15	Environmental	Cerro	11/5/08	PAN0	N	
1097	LR-1305-02	FSL R8-2847	15	Environmental	Cerro	11/5/08	PAN0	Y	CU-213
1116	LR-1318-01	FSL R8-2866	15	Environmental	Cerro	12/9/08	PAN0	N	
1117	LR-1318-02	FSL R8-2867	15	Environmental	Cerro	12/9/08	PAN0	Y	CU-213
139	LR-1063-01	FSL R8-061	16	Environmental	8,20:-:z6	12/4/07	PAN0	Y	CU-96
303	LR-1121-01	FSL R8-836	16	Bov non-clin	Kentucky	2/13/08	PAN0	Y	CU-96
304	LR-1121-02	FSL R8-837	16	Bov non-clin	Kentucky	2/13/08	PAN0	N	
305	LR-1122-01	FSL R8-838	16	Bov non-clin	Kentucky	2/13/08	CHL2	Y	CU-96
350	LR-1139-01	FSL R8-900	16	Environmental	Kentucky	4/3/08	AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-184
417	LR-1159-01	FSL R8-967	16	Bov non-clin	Kentucky	4/3/08	PAN0	Y	CU-96
612	LR-1194-01	FSL R8-1485	16	Bov non-clin	Kentucky* (Untypeable Rough O:i:z6)	5/21/08	PAN0	Y	CU-96
787	LR-1241-01	FSL R8-2031	16	Environmental	Kentucky* (Untypeable Rough O:NonMotile)	9/9/08	PAN0	Y	CU-96
1098	LR-1306-01	FSL R8-2848	16	Environmental	Kentucky	11/11/08	PAN0	Y	CU-96
38	LR-1025-01	FSL R6-1000	17	Environmental	Kentucky (Newport)	10/11/07	AMC1 AMP1 FOX1 CEF1 CRO1 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-96

Appendix Table 1

68	LR-1039-01	FSL R6-956	17	Bov non-clin	Newport	11/7/07	AMC1 AMP1 FOX1 CEF1 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
69	LR-1040-01	FSL R6-957	17	Bov non-clin	Kentucky	11/7/07	PAN0	N	
70	LR-1040-02	FSL R6-958	17	Bov non-clin	Kentucky	11/7/07	PAN0	Y	CU-96
71	LR-1040-03	FSL R6-959	17	Bov non-clin	Kentucky	11/7/07	PAN0	N	
211	LR-1088-01	FSL R8-206	17	Bov non-clin	Newport	12/12/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
212	LR-1088-02	FSL R8-207	17	Bov non-clin	Newport	12/12/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
213	LR-1089-01	FSL R8-208	17	Bov non-clin	Typhimurium* (Cerro)	12/12/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-71
214	LR-1088-03	FSL R8-209	17	Bov non-clin	Newport	12/12/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
215	LR-1088-04	FSL R8-210	17	Bov non-clin	Newport	12/12/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
216	LR-1090-01	FSL R8-211	17	Bov non-clin	Kentucky	12/12/07	PAN0	N	
217	LR-1090-02	FSL R8-212	17	Bov non-clin	Kentucky	12/12/07	PAN0	Y	CU-96
218	LR-1090-03	FSL R8-213	17	Bov non-clin	Kentucky	12/12/07	PAN0	N	
219	LR-1090-04	FSL R8-214	17	Bov non-clin	Kentucky	12/12/07	PAN0	N	
220	LR-1088-05	FSL R8-215	17	Bov non-clin	Newport	12/12/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
102	LR-1052-01	FSL R8-341	17	Environmental	Newport	2/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
103	LR-1053-01	FSL R8-342	17	Environmental	Newport (Meleagridis)	2/12/08	FOX1 CHL2	Y	CU-121
290	LR-1116-01	FSL R8-823	17	Bov non-clin	Kentucky	2/12/08	PAN0	N	
291	LR-1116-02	FSL R8-824	17	Bov non-clin	Kentucky	2/12/08	PAN0	Y	CU-96
292	LR-1116-03	FSL R8-825	17	Bov non-clin	Kentucky	2/12/08	PAN0	N	
293	LR-1116-04	FSL R8-826	17	Bov non-clin	Kentucky	2/12/08	PAN0	N	
294	LR-1116-05	FSL R8-827	17	Bov non-clin	Kentucky	2/12/08	PAN0	N	
295	LR-1117-01	FSL R8-828	17	Bov non-clin	Newport	2/12/08	AMC1 AMP1 FOX1 CEF1 CRO1 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
296	LR-1118-01	FSL R8-829	17	Bov non-clin	Newport	2/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
297	LR-1118-02	FSL R8-830	17	Bov non-clin	Newport (Kentucky)	2/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	E	
298	LR-1118-03	FSL R8-831	17	Bov non-clin	Newport	2/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
281	LR-1110-01	FSL R8-814	17	Environmental	Kentucky	3/11/08	PAN0	Y	CU-96
282	LR-1111-01	FSL R8-815	17	Environmental	Kentucky	3/11/08	TEL1	Y	CU-834
359	LR-1145-01	FSL R8-909	17	Environmental	Cerro	4/22/08	PAN0	N	
360	LR-1145-02	FSL R8-910	17	Environmental	Cerro	4/22/08	PAN0	Y	CU-213
361	LR-1145-03	FSL R8-911	17	Environmental	Cerro	4/22/08	PAN0	N	
532	LR-1181-01	FSL R8-1405	17	Environmental	Kentucky	6/17/08	PAN0	Y	CU-96
632	LR-1208-01	FSL R8-1641	17	Environmental	Kentucky	7/27/08	PAN0	Y	CU-96
788	LR-1242-01	FSL R8-2032	17	Environmental	Kentucky	9/10/08	PAN0	N	
789	LR-1242-02	FSL R8-2033	17	Environmental	Kentucky	9/10/08	PAN0	Y	CU-96
16	LR-1012-01	FSL R6-789	18	Environmental	Infantis	10/18/07	PAN0	N	
17	LR-1012-02	FSL R6-790	18	Environmental	Infantis	10/18/07	PAN0	N	
18	LR-1012-03	FSL R6-791	18	Environmental	Infantis	10/18/07	PAN0	Y	CU-107
72	LR-1041-01	FSL R6-960	18	Bov non-clin	Newport	11/8/07	PAN0	N	
73	LR-1041-02	FSL R6-961	18	Bov non-clin	Newport	11/8/07	PAN0	Y	CU-328
221	LR-1091-01	FSL R8-216	18	Bov non-clin	Newport	1/11/08	PAN0	Y	CU-328
336	LR-1133-01	FSL R8-886	18	Environmental	Cerro	3/19/08	PAN0	Y	CU-213
337	LR-1133-02	FSL R8-887	18	Environmental	Cerro	3/19/08	PAN0	N	
362	LR-1146-01	FSL R8-912	18	Environmental	Kentucky	4/21/08	PAN0	Y	CU-96
520	LR-1174-01	FSL R8-1393	18	Environmental	Cerro	5/28/08	PAN0	Y	CU-213
521	LR-1174-02	FSL R8-1394	18	Environmental	Cerro	5/28/08	PAN0	N	
545	LR-1190-01	FSL R8-1418	18	Environmental	Cerro (Oranienburg)	6/30/08	PAN0	E	
546	LR-1190-02	FSL R8-1419	18	Environmental	Cerro	6/30/08	PAN0	N	
547	LR-1190-03	FSL R8-1420	18	Environmental	Cerro	6/30/08	PAN0	Y	CU-213
784	LR-1239-01	FSL R8-2028	18	Environmental	Cerro	9/8/08	PAN0	N	
785	LR-1239-02	FSL R8-2029	18	Environmental	Cerro	9/8/08	PAN0	Y	CU-213
12	LR-1009-01	FSL R6-785	19	Environmental	Typhimurium	10/18/07	PAN0	Y	CU-914
13	LR-1010-01	FSL R6-786	19	Environmental	Typhimurium Copenhagen	10/18/07	PAN0	Y	CU-914
14	LR-1009-02	FSL R6-787	19	Environmental	Typhimurium	10/18/07	PAN0	N	
15	LR-1011-01	FSL R6-788	19	Environmental	Anatum	10/18/07	PAN0	Y	CU-431
74	LR-1042-01	FSL R6-962	19	Bov non-clin	Anatum* (Untypeable)	11/19/07	PAN0	Y	CU-431
75	LR-1043-01	FSL R6-963	19	Bov non-clin	Anatum	11/19/07	PAN0	Y	CU-431
76	LR-1043-02	FSL R6-964	19	Bov non-clin	Anatum	11/19/07	PAN0	N	
77	LR-1044-01	FSL R6-965	19	Bov non-clin	Newport	11/19/07	PAN0	Y	CU-1015
78	LR-1045-01	FSL R6-966	19	Bov non-clin	Kentucky	11/19/07	PAN0	N	

Appendix Table 1

79	LR-1043-03	FSL R6-967	19	Bov non-clin	Anatum	11/19/07	PAN0	N	
80	LR-1045-02	FSL R6-968	19	Bov non-clin	Kentucky	11/19/07	PAN0	Y	CU-96
81	LR-1043-04	FSL R6-969	19	Bov non-clin	Anatum	11/19/07	PAN0	N	
82	LR-1045-03	FSL R6-970	19	Bov non-clin	Kentucky	11/19/07	PAN0	N	
83	LR-1043-05	FSL R6-971	19	Bov non-clin	Anatum	11/19/07	PAN0	N	
84	LR-1045-04	FSL R6-972	19	Bov non-clin	Kentucky	11/19/07	PAN0	N	
85	LR-1045-05	FSL R6-973	19	Bov non-clin	Kentucky	11/19/07	PAN0	N	
86	LR-1043-06	FSL R6-974	19	Bov non-clin	Anatum	11/19/07	PAN0	N	
87	LR-1043-07	FSL R6-975	19	Bov non-clin	Anatum	11/19/07	PAN0	N	
173	LR-1072-01	FSL R8-168	19	Environmental	Kentucky	1/16/08	PAN0	Y	CU-96
174	LR-1073-01	FSL R8-169	19	Environmental	Oranienburg	1/16/08	PAN0	Y	CU-040
175	LR-1073-02	FSL R8-170	19	Environmental	Oranienburg	1/16/08	PAN0	N	
222	LR-1092-01	FSL R8-217	19	Bov non-clin	Kentucky	1/16/08	PAN0	N	
223	LR-1092-02	FSL R8-218	19	Bov non-clin	Kentucky	1/16/08	PAN0	N	
224	LR-1092-03	FSL R8-219	19	Bov non-clin	Kentucky	1/16/08	PAN0	N	
225	LR-1092-04	FSL R8-220	19	Bov non-clin	Kentucky	1/16/08	PAN0	Y	CU-96
226	LR-1092-05	FSL R8-221	19	Bov non-clin	Kentucky	1/16/08	PAN0	N	
227	LR-1092-06	FSL R8-222	19	Bov non-clin	Kentucky	1/16/08	PAN0	N	
283	LR-1112-01	FSL R8-816	19	Environmental	Kentucky	3/13/08	PAN0	N	
284	LR-1112-02	FSL R8-817	19	Environmental	Kentucky	3/13/08	PAN0	Y	CU-96
285	LR-1112-03	FSL R8-818	19	Environmental	Kentucky	3/13/08	PAN0	N	
323	LR-1128-01	FSL R8-856	19	Bov non-clin	Kentucky	3/13/08	PAN0	N	
324	LR-1128-02	FSL R8-857	19	Bov non-clin	Kentucky	3/13/08	PAN0	N	
325	LR-1128-03	FSL R8-858	19	Bov non-clin	Kentucky	3/13/08	PAN0	N	
326	LR-1128-04	FSL R8-859	19	Bov non-clin	Kentucky	3/13/08	PAN0	Y	CU-96
327	LR-1128-05	FSL R8-860	19	Bov non-clin	Kentucky	3/13/08	PAN0	N	
328	LR-1128-06	FSL R8-861	19	Bov non-clin	Kentucky	3/13/08	PAN0	N	
329	LR-1128-07	FSL R8-862	19	Bov non-clin	Kentucky	3/13/08	PAN0	N	
366	LR-1150-01	FSL R8-916	19	Environmental	Kentucky	4/28/08	PAN0	Y	CU-96
367	LR-1151-01	FSL R8-917	19	Environmental	Kentucky	4/28/08	AMC1 AMP1 FOX1	Y	CU-96
635	LR-1210-01	FSL R8-1644	19	Environmental	Kentucky	7/21/08	PAN0	Y	CU-96
636	LR-1210-02	FSL R8-1645	19	Environmental	Kentucky	7/21/08	PAN0	N	
637	LR-1211-01	FSL R8-1646	19	Environmental	6,7:-:1,5	7/21/08	PAN0	Y	CU-1027
638	LR-1210-03	FSL R8-1647	19	Environmental	Kentucky	7/21/08	PAN0	N	
1063	LR-1289-01	FSL R8-2813	19	Environmental	Kentucky	10/2/08	PAN0	N	
1064	LR-1289-02	FSL R8-2814	19	Environmental	Kentucky	10/2/08	PAN0	Y	CU-96
1065	LR-1289-03	FSL R8-2815	19	Environmental	Kentucky	10/2/08	PAN0	N	
1066	LR-1289-04	FSL R8-2816	19	Environmental	Kentucky	10/2/08	PAN0	N	
530	LR-1179-01	FSL R8-1403	20	Environmental	Paratyphi B var. L-tartrate+	6/10/08	PAN0	Y	CU-187
22	LR-1016-01	FSL R6-795	21	Environmental	Kentucky	10/19/07	PAN0	N	
23	LR-1016-02	FSL R6-796	21	Environmental	Kentucky	10/19/07	PAN0	Y	CU-96
88	LR-1046-01	FSL R6-976	21	Bov non-clin	Kentucky	11/19/07	PAN0	Y	CU-96
114	LR-1059-01	FSL R8-353	21	Environmental	Anatum	3/6/08	PAN0	N	
115	LR-1059-02	FSL R8-354	21	Environmental	Anatum	3/6/08	PAN0	Y	CU-431
322	LR-1127-01	FSL R8-855	21	Bov non-clin	Anatum	3/6/08	PAN0	Y	CU-431
531	LR-1180-01	FSL R8-1404	21	Environmental	Anatum	6/16/08	PAN0	Y	CU-431
176	LR-1074-01	FSL R8-171	22	Environmental	Typhimurium	12/17/07	PAN0	Y	CU-71
177	LR-1075-01	FSL R8-172	22	Environmental	Typhimurium	1/18/08	PAN0	N	
178	LR-1075-02	FSL R8-173	22	Environmental	Typhimurium	1/18/08	PAN0	N	
179	LR-1075-03	FSL R8-174	22	Environmental	Typhimurium	1/18/08	PAN0	Y	CU-71
180	LR-1075-04	FSL R8-175	22	Environmental	Typhimurium	1/18/08	PAN0	N	
228	LR-1093-01	FSL R8-223	22	Bov non-clin	Typhimurium	1/18/08	PAN0	Y	CU-71
229	LR-1093-02	FSL R8-224	22	Bov non-clin	Typhimurium	1/18/08	PAN0	N	
230	LR-1093-03	FSL R8-225	22	Bov non-clin	Typhimurium	1/18/08	PAN0	N	
231	LR-1093-04	FSL R8-226	22	Bov non-clin	Typhimurium	1/18/08	PAN0	N	
232	LR-1094-01	FSL R8-227	22	Bov non-clin	Typhimurium	1/18/08	AMC1 AMP1 FOX2 CEF1 CHL2 SUL1	Y	CU-71
233	LR-1095-01	FSL R8-228	22	Bov non-clin	Typhimurium	1/18/08	CHL2 TEL1	Y	CU-71
234	LR-1096-01	FSL R8-229	22	Bov non-clin	Typhimurium	1/18/08	AMC1 AMP1 CHL2 SUL1	Y	CU-71

Appendix Table 1

235	LR-1093-05	FSL R8-230	22	Bov non-clin	Typhimurium	1/18/08	PAN0	N	
236	LR-1097-01	FSL R8-231	22	Bov non-clin	Typhimurium	1/18/08	AMC1 AMP1 CEF1 CHL2 SUL1	Y	CU-71
237	LR-1093-06	FSL R8-232	22	Bov non-clin	Typhimurium	1/18/08	PAN0	N	
276	LR-1107-01	FSL R8-271	22	Bov non-clin	Typhimurium	1/18/08	AMC1 AMP1 FOX1 CEF1 CRO1 CHL1 NAL1 SUL1 TEL1	Y	CU-71
277	LR-1093-07	FSL R8-272	22	Bov non-clin	Typhimurium	1/18/08	PAN0	N	
278	LR-1108-01	FSL R8-273	22	Bov non-clin	Typhimurium	1/18/08	AMC1 AMP1 FOX2 CEF1 SUL1 TEL1	Y	CU-71
286	LR-1113-01	FSL R8-819	22	Environmental	Typhimurium	3/14/08	PAN0	N	
287	LR-1113-02	FSL R8-820	22	Environmental	Typhimurium	3/14/08	PAN0	Y	CU-71
332	LR-1130-01	FSL R8-865	22	Bov non-clin	Typhimurium	3/14/08	AMP1 CEF1 CRO1	Y	CU-71
333	LR-1131-01	FSL R8-866	22	Bov non-clin	Typhimurium	3/14/08	PAN0	N	
334	LR-1131-02	FSL R8-867	22	Bov non-clin	Typhimurium	3/14/08	PAN0	Y	CU-71
24	LR-1017-01	FSL R6-797	23	Environmental	Muenster	10/19/07	PAN0	Y	CU-170
19	LR-1013-01	FSL R6-792	25	Environmental	Typhimurium	10/19/07	FOX1 CEF2 CHL1	Y	CU-67
20	LR-1014-01	FSL R6-793	25	Environmental	Typhimurium	10/19/07	AMC1 FOX1 CEF2 CHL1	Y	CU-67
21	LR-1015-01	FSL R6-794	25	Environmental	Typhimurium	10/19/07	FOX1 CEF1	Y	CU-67
89	LR-1047-01	FSL R6-977	25	Bov non-clin	Typhimurium	11/20/07	PAN0	N	
90	LR-1048-01	FSL R6-978	25	Bov non-clin	Typhimurium	11/20/07	AMC1 AMP1 FOX1 CEF1	Y	CU-67
91	LR-1049-01	FSL R6-979	25	Bov non-clin	Typhimurium Copenhagen	11/20/07	FOX1 CHL1	Y	CU-67
92	LR-1048-02	FSL R6-980	25	Bov non-clin	Typhimurium	11/20/07	AMC1 AMP1 FOX1 CEF1	N	
93	LR-1048-03	FSL R6-981	25	Bov non-clin	Typhimurium	11/20/07	AMC1 AMP1 FOX1 CEF1	N	
94	LR-1048-04	FSL R6-982	25	Bov non-clin	Typhimurium	11/20/07	AMC1 AMP1 FOX1 CEF1	N	
95	LR-1047-02	FSL R6-983	25	Bov non-clin	Typhimurium	11/20/07	PAN0	Y	CU-67
96	LR-1047-03	FSL R6-984	25	Bov non-clin	Typhimurium	11/20/07	PAN0	N	
181	LR-1076-01	FSL R8-176	25	Environmental	Typhimurium	1/15/08	CEF1 CRO2	N	
182	LR-1076-02	FSL R8-177	25	Environmental	Typhimurium	1/15/08	CEF1 CRO2	Y	CU-67
183	LR-1076-03	FSL R8-178	25	Environmental	Typhimurium	1/15/08	CEF1 CRO2	N	
238	LR-1098-01	FSL R8-233	25	Bov non-clin	Typhimurium	1/15/08	CEF1 CRO2	Y	CU-67
279	LR-1109-01	FSL R8-812	25	Environmental	Typhimurium	3/7/08	AMC1 AMP1 FOX1 CEF1 CRO2	Y	CU-67
280	LR-1109-02	FSL R8-813	25	Environmental	Typhimurium	3/7/08	AMC1 AMP1 FOX1 CEF1 CRO2	N	
346	LR-1137-01	FSL R8-896	25	Environmental	Typhimurium Copenhagen	4/1/08	AMC2 AMP1 FOX1 CEF2 KAN1 STR1 SUL1 TEL1	Y	CU-104
347	LR-1138-01	FSL R8-897	25	Environmental	Typhimurium Copenhagen	4/1/08	AMP1 KAN1 STR1 SUL1 TEL1	N	
348	LR-1138-02	FSL R8-898	25	Environmental	Typhimurium Copenhagen	4/1/08	AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-104
349	LR-1138-03	FSL R8-899	25	Environmental	Typhimurium Copenhagen	4/1/08	AMP1 KAN1 STR1 SUL1 TEL1	N	
512	LR-1171-01	FSL R8-1385	25	Environmental	Typhimurium	5/1/08	AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-104
513	LR-1171-02	FSL R8-1386	25	Environmental	Typhimurium	5/1/08	AMP1 KAN1 STR1 SUL1 TEL1	N	
514	LR-1171-03	FSL R8-1387	25	Environmental	Typhimurium	5/1/08	AMP1 KAN1 STR1 SUL1 TEL1	N	
613	LR-1195-01	FSL R8-1621	25	Environmental	Typhimurium	7/1/08	AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-1034
614	LR-1196-01	FSL R8-1622	25	Environmental	Typhimurium	7/1/08	AMC2 AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-1034
615	LR-1197-01	FSL R8-1623	25	Environmental	Typhimurium Copenhagen	7/1/08	AMC1 AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-104
633	LR-1209-01	FSL R8-1642	25	Environmental	Typhimurium Copenhagen	7/29/08	AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-1031
634	LR-1209-02	FSL R8-1643	25	Environmental	Typhimurium Copenhagen	7/29/08	AMP1 KAN1 STR1 SUL1 TEL1	N	
774	LR-1232-01	FSL R8-2017	25	Environmental	Typhimurium Copenhagen	9/3/08	AMP1 KAN1 STR1 SUL1 TEL1	Y	CU-104
5	LR-1004-01	FSL R6-778	26	Environmental	Muenster	10/24/07	AMC1 AMP1 FOX1 CEF1 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-173
6	LR-1005-01	FSL R6-779	26	Environmental	Kentucky (Cerro)	10/24/07	PAN0	Y	CU-96
7	LR-1006-01	FSL R6-780	26	Environmental	Muenster	10/24/07	PAN0	Y	CU-173
140	LR-1064-01	FSL R8-062	26	Bov non-clin	Muenster	11/27/07	PAN0	Y	CU-173
141	LR-1065-01	FSL R8-063	26	Bov non-clin	4,5,12:i:-	11/27/07	PAN0	Y	CU-89
142	LR-1066-01	FSL R8-064	26	Bov non-clin	Kentucky	11/27/07	PAN0	N	
143	LR-1066-02	FSL R8-065	26	Bov non-clin	Kentucky	11/27/07	PAN0	Y	CU-96
144	LR-1067-01	FSL R8-066	26	Bov non-clin	Cerro	11/27/07	PAN0	N	
145	LR-1066-03	FSL R8-067	26	Bov non-clin	Kentucky	11/27/07	PAN0	N	
146	LR-1066-04	FSL R8-068	26	Bov non-clin	Kentucky	11/27/07	PAN0	N	
147	LR-1066-05	FSL R8-069	26	Bov non-clin	Kentucky	11/27/07	PAN0	N	
148	LR-1064-02	FSL R8-070	26	Bov non-clin	Muenster	11/27/07	PAN0	N	
149	LR-1064-03	FSL R8-071	26	Bov non-clin	Muenster	11/27/07	PAN0	N	
150	LR-1067-02	FSL R8-072	26	Bov non-clin	Cerro	11/27/07	PAN0	Y	CU-213
151	LR-1066-06	FSL R8-073	26	Bov non-clin	Kentucky	11/27/07	PAN0	N	
152	LR-1064-04	FSL R8-074	26	Bov non-clin	Muenster	11/27/07	PAN0	N	
153	LR-1064-05	FSL R8-075	26	Bov non-clin	Muenster	11/27/07	PAN0	N	
184	LR-1077-01	FSL R8-179	26	Environmental	Muenster	1/24/08	PAN0	Y	CU-173

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185	LR-1078-01	FSL R8-180	26	Environmental	Kentucky	1/24/08	PAN0	Y	CU-96
186	LR-1078-02	FSL R8-181	26	Environmental	Kentucky	1/24/08	PAN0	N	
187	LR-1079-01	FSL R8-182	26	Environmental	Cerro	1/24/08	PAN0	Y	CU-213
239	LR-1099-01	FSL R8-234	26	Bov non-clin	Cerro	1/24/08	PAN0	N	
240	LR-1099-02	FSL R8-235	26	Bov non-clin	Cerro	1/24/08	PAN0	Y	CU-213
241	LR-1099-03	FSL R8-236	26	Bov non-clin	Cerro	1/24/08	PAN0	N	
242	LR-1099-04	FSL R8-237	26	Bov non-clin	Cerro	1/24/08	PAN0	N	
243	LR-1100-01	FSL R8-238	26	Bov non-clin	Kentucky	1/24/08	PAN0	Y	CU-96
244	LR-1099-05	FSL R8-239	26	Bov non-clin	Cerro	1/24/08	PAN0	N	
245	LR-1100-02	FSL R8-240	26	Bov non-clin	Kentucky	1/24/08	PAN0	N	
246	LR-1099-06	FSL R8-241	26	Bov non-clin	Cerro	1/24/08	PAN0	N	
1256	LR-1351-01	FSL R8-3414	26	Environmental	Kentucky	1/26/08	PAN0	N	
1257	LR-1351-02	FSL R8-3415	26	Environmental	Kentucky	1/26/08	PAN0	Y	CU-96
1258	LR-1352-01	FSL R8-3416	26	Environmental	Cerro	1/26/08	PAN0	Y	CU-213
335	LR-1132-01	FSL R8-885	26	Environmental	Cerro	3/19/08	PAN0	Y	CU-213
371	LR-1154-01	FSL R8-921	26	Bov non-clin	Cerro	3/19/08	PAN0	N	
372	LR-1155-01	FSL R8-922	26	Bov non-clin	Kentucky	3/19/08	PAN0	Y	CU-96
373	LR-1154-02	FSL R8-923	26	Bov non-clin	Cerro	3/19/08	PAN0	N	
374	LR-1154-03	FSL R8-924	26	Bov non-clin	Cerro	3/19/08	PAN0	N	
375	LR-1154-04	FSL R8-925	26	Bov non-clin	Cerro	3/19/08	PAN0	N	
376	LR-1155-02	FSL R8-926	26	Bov non-clin	Kentucky	3/19/08	PAN0	N	
377	LR-1154-05	FSL R8-927	26	Bov non-clin	Cerro	3/19/08	PAN0	N	
378	LR-1156-01	FSL R8-928	26	Bov non-clin	Kentucky	3/19/08	TEL1	Y	CU-96
379	LR-1154-06	FSL R8-929	26	Bov non-clin	Cerro	3/19/08	PAN0	Y	CU-213
380	LR-1155-03	FSL R8-930	26	Bov non-clin	Kentucky	3/19/08	PAN0	N	
522	LR-1175-01	FSL R8-1395	26	Environmental	Cerro	6/2/08	PAN0	Y	CU-213
523	LR-1175-02	FSL R8-1396	26	Environmental	Cerro (Kentucky)	6/2/08	PAN0	E	
616	LR-1198-01	FSL R8-1624	26	Environmental	Cerro	7/7/08	PAN0	Y	CU-213
795	LR-1246-01	FSL R8-2039	26	Environmental	Cerro	9/10/08	PAN0	N	
796	LR-1247-01	FSL R8-2040	26	Environmental	Kentucky	9/10/08	PAN0	Y	CU-96
797	LR-1246-02	FSL R8-2041	26	Environmental	Cerro	9/10/08	PAN0	Y	CU-213
1081	LR-1299-01	FSL R8-2831	26	Environmental	Kentucky	10/27/08	PAN0	N	
1082	LR-1299-02	FSL R8-2832	26	Environmental	Kentucky	10/27/08	PAN0	Y	CU-96
1083	LR-1299-03	FSL R8-2833	26	Environmental	Kentucky	10/27/08	PAN0	N	
11	LR-1008-01	FSL R6-784	27	Environmental	Cerro	10/24/07	PAN0	Y	CU-213
188	LR-1080-01	FSL R8-183	27	Environmental	Cerro	1/25/08	PAN0	N	
189	LR-1081-01	FSL R8-184	27	Environmental	Cerro	1/25/08	AMC1 AMP1 TEL1	Y	CU-213
190	LR-1080-02	FSL R8-185	27	Environmental	Cerro	1/25/08	PAN0	Y	CU-213
1	LR-1001-01	FSL R6-774	28	Environmental	Agona	10/24/07	PAN0	Y	CU-812
2	LR-1002-01	FSL R6-775	28	Environmental	Cerro	10/24/07	PAN0	Y	CU-213
3	LR-1003-01	FSL R6-776	29	Environmental	Newport	10/24/07	AMC1 AMP1 FOX1 CEF1 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
4	LR-1003-02	FSL R6-777	29	Environmental	Newport	10/24/07	AMC1 AMP1 FOX1 CEF1 CHL1 KAN1 STR1 SUL1 TEL1	N	
8	LR-1007-01	FSL R6-781	30	Environmental	Cerro	10/24/07	PAN0	Y	CU-213
9	LR-1007-02	FSL R6-782	30	Environmental	Cerro (Multiple serovars)	10/24/07	PAN0	E	
10	LR-1007-03	FSL R6-783	30	Environmental	Cerro	10/24/07	PAN0	N	
154	LR-1068-01	FSL R8-076	30	Bov non-clin	Cerro	11/28/07	PAN0	Y	CU-213
155	LR-1068-02	FSL R8-077	30	Bov non-clin	Cerro	11/28/07	PAN0	N	
156	LR-1068-03	FSL R8-078	30	Bov non-clin	Cerro	11/28/07	PAN0	N	
157	LR-1068-04	FSL R8-079	30	Bov non-clin	Cerro	11/28/07	PAN0	N	
158	LR-1068-05	FSL R8-080	30	Bov non-clin	Cerro	11/28/07	PAN0	N	
159	LR-1068-06	FSL R8-081	30	Bov non-clin	Cerro	11/28/07	PAN0	N	
160	LR-1068-07	FSL R8-082	30	Bov non-clin	Cerro	11/28/07	PAN0	N	
191	LR-1082-01	FSL R8-186	30	Environmental	Cerro	1/23/08	PAN0	Y	CU-213
247	LR-1101-01	FSL R8-242	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
248	LR-1101-02	FSL R8-243	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
249	LR-1101-03	FSL R8-244	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
250	LR-1101-04	FSL R8-245	30	Bov non-clin	Cerro	1/23/08	PAN0	Y	CU-213
251	LR-1101-05	FSL R8-246	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
252	LR-1102-01	FSL R8-247	30	Bov non-clin	Cerro	1/23/08	SUL1	Y	CU-213

Appendix Table 1

253	LR-1101-06	FSL R8-248	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
254	LR-1101-07	FSL R8-249	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
255	LR-1101-08	FSL R8-250	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
256	LR-1103-01	FSL R8-251	30	Bov non-clin	Cerro	1/23/08	AMC2 AMP1 CEF1 SUL1	Y	CU-841
257	LR-1101-09	FSL R8-252	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
258	LR-1101-10	FSL R8-253	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
259	LR-1104-01	FSL R8-254	30	Bov non-clin	Cerro	1/23/08	AMC2 SUL1	Y	CU-213
260	LR-1101-11	FSL R8-255	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
261	LR-1101-12	FSL R8-256	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
262	LR-1101-13	FSL R8-257	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
263	LR-1101-14	FSL R8-258	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
264	LR-1101-15	FSL R8-259	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
265	LR-1101-16	FSL R8-260	30	Bov non-clin	Cerro	1/23/08	PAN0	N	
450	LR-1165-01	FSL R8-1000	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
451	LR-1165-02	FSL R8-1001	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
452	LR-1165-03	FSL R8-1002	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
453	LR-1165-04	FSL R8-1003	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
454	LR-1165-05	FSL R8-1004	30	Bov non-clin	Cerro	3/18/08	PAN0	Y	CU-213
455	LR-1165-06	FSL R8-1005	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
456	LR-1165-07	FSL R8-1006	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
457	LR-1165-08	FSL R8-1007	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
458	LR-1165-09	FSL R8-1008	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
459	LR-1165-10	FSL R8-1009	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
460	LR-1165-11	FSL R8-1010	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
461	LR-1165-12	FSL R8-1011	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
462	LR-1165-13	FSL R8-1012	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
463	LR-1165-14	FSL R8-1013	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
464	LR-1165-15	FSL R8-1014	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
465	LR-1165-16	FSL R8-1015	30	Bov non-clin	Cerro	3/18/08	PAN0	N	
341	LR-1135-01	FSL R8-891	30	Environmental	Cerro	3/25/08	PAN0	Y	CU-213
342	LR-1135-02	FSL R8-892	30	Environmental	Cerro	3/25/08	PAN0	N	
381	LR-1157-01	FSL R8-931	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
382	LR-1157-02	FSL R8-932	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
383	LR-1157-03	FSL R8-933	30	Bov non-clin	Cerro	3/25/08	PAN0	Y	CU-213
384	LR-1157-04	FSL R8-934	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
385	LR-1157-05	FSL R8-935	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
386	LR-1157-06	FSL R8-936	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
387	LR-1157-07	FSL R8-937	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
388	LR-1157-08	FSL R8-938	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
389	LR-1157-09	FSL R8-939	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
390	LR-1157-10	FSL R8-940	30	Bov non-clin	Cerro	3/25/08	PAN0	N	
466	LR-1166-01	FSL R8-1016	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
467	LR-1166-02	FSL R8-1017	30	Bov non-clin	Cerro	4/1/08	PAN0	Y	CU-213
468	LR-1166-03	FSL R8-1018	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
469	LR-1166-04	FSL R8-1019	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
470	LR-1166-05	FSL R8-1020	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
471	LR-1166-06	FSL R8-1021	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
472	LR-1166-07	FSL R8-1022	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
473	LR-1166-08	FSL R8-1023	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
474	LR-1166-09	FSL R8-1024	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
475	LR-1166-10	FSL R8-1025	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
476	LR-1166-11	FSL R8-1026	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
477	LR-1166-12	FSL R8-1027	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
478	LR-1166-13	FSL R8-1028	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
479	LR-1166-14	FSL R8-1029	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
480	LR-1166-15	FSL R8-1030	30	Bov non-clin	Cerro	4/1/08	PAN0	N	

Appendix Table 1

481	LR-1166-16	FSL R8-1031	30	Bov non-clin	Cerro	4/1/08	PAN0	N	
482	LR-1167-01	FSL R8-1032	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
483	LR-1167-02	FSL R8-1033	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
484	LR-1167-03	FSL R8-1034	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
485	LR-1167-04	FSL R8-1035	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
486	LR-1167-05	FSL R8-1036	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
487	LR-1167-06	FSL R8-1037	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
488	LR-1167-07	FSL R8-1038	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
489	LR-1167-08	FSL R8-1039	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
490	LR-1167-09	FSL R8-1040	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
491	LR-1167-10	FSL R8-1041	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
492	LR-1167-11	FSL R8-1042	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
493	LR-1167-12	FSL R8-1043	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
494	LR-1167-13	FSL R8-1044	30	Bov non-clin	Cerro	4/16/08	PAN0	Y	CU-841
495	LR-1167-14	FSL R8-1045	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
496	LR-1167-15	FSL R8-1046	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
497	LR-1167-16	FSL R8-1047	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
498	LR-1167-17	FSL R8-1048	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
499	LR-1167-18	FSL R8-1049	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
500	LR-1167-19	FSL R8-1050	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
501	LR-1167-20	FSL R8-1051	30	Bov non-clin	Cerro	4/16/08	PAN0	N	
368	LR-1152-01	FSL R8-918	30	Environmental	Cerro	4/28/08	PAN0	Y	CU-213
369	LR-1152-02	FSL R8-919	30	Environmental	Cerro	4/28/08	PAN0	N	
502	LR-1168-01	FSL R8-1052	30	Bov non-clin	Cerro	4/28/08	PAN0	N	
503	LR-1168-02	FSL R8-1053	30	Bov non-clin	Cerro	4/28/08	PAN0	N	
504	LR-1168-03	FSL R8-1054	30	Bov non-clin	Cerro	4/28/08	PAN0	N	
505	LR-1168-04	FSL R8-1055	30	Bov non-clin	Cerro	4/28/08	PAN0	N	
506	LR-1168-05	FSL R8-1056	30	Bov non-clin	Cerro	4/28/08	PAN0	N	
507	LR-1168-06	FSL R8-1057	30	Bov non-clin	Cerro	4/28/08	PAN0	Y	CU-213
508	LR-1168-07	FSL R8-1058	30	Bov non-clin	Cerro	4/28/08	PAN0	N	
677	LR-1215-01	FSL R8-1686	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
678	LR-1215-02	FSL R8-1687	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
679	LR-1215-03	FSL R8-1688	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
680	LR-1215-04	FSL R8-1689	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
681	LR-1215-05	FSL R8-1690	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
682	LR-1215-06	FSL R8-1691	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
683	LR-1215-07	FSL R8-1692	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
684	LR-1215-08	FSL R8-1693	30	Bov non-clin	Cerro	5/12/08	PAN0	Y	CU-213
685	LR-1215-09	FSL R8-1694	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
686	LR-1215-10	FSL R8-1695	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
687	LR-1215-11	FSL R8-1696	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
688	LR-1215-12	FSL R8-1697	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
689	LR-1215-13	FSL R8-1698	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
690	LR-1215-14	FSL R8-1699	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
691	LR-1215-15	FSL R8-1700	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
692	LR-1215-16	FSL R8-1701	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
693	LR-1216-01	FSL R8-1702	30	Bov non-clin	Cerro	5/12/08	AMC1 AMP2 TEL1	Y	CU-213
694	LR-1217-01	FSL R8-1703	30	Bov non-clin	Cerro	5/12/08	AMC1 AMP1 TEL1	Y	CU-213
695	LR-1215-17	FSL R8-1704	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
696	LR-1215-18	FSL R8-1705	30	Bov non-clin	Cerro	5/12/08	PAN0	N	
697	LR-1218-01	FSL R8-1706	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
698	LR-1218-02	FSL R8-1707	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
699	LR-1218-03	FSL R8-1708	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
700	LR-1218-04	FSL R8-1709	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
701	LR-1218-05	FSL R8-1710	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
702	LR-1218-06	FSL R8-1711	30	Bov non-clin	Cerro	5/26/08	PAN0	N	

Appendix Table 1

703	LR-1218-07	FSL R8-1712	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
704	LR-1218-08	FSL R8-1713	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
705	LR-1218-09	FSL R8-1714	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
706	LR-1218-10	FSL R8-1715	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
707	LR-1218-11	FSL R8-1716	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
708	LR-1218-12	FSL R8-1717	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
709	LR-1218-13	FSL R8-1718	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
710	LR-1218-14	FSL R8-1719	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
711	LR-1218-15	FSL R8-1720	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
712	LR-1218-16	FSL R8-1721	30	Bov non-clin	Cerro	5/26/08	PAN0	Y	CU-841
713	LR-1218-17	FSL R8-1722	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
714	LR-1218-18	FSL R8-1723	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
715	LR-1218-19	FSL R8-1724	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
716	LR-1218-20	FSL R8-1725	30	Bov non-clin	Cerro	5/26/08	PAN0	N	
733	LR-1220-01	FSL R8-1742	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
734	LR-1220-02	FSL R8-1743	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
735	LR-1220-03	FSL R8-1744	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
736	LR-1220-04	FSL R8-1745	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
737	LR-1220-05	FSL R8-1746	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
738	LR-1220-06	FSL R8-1747	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
739	LR-1220-07	FSL R8-1748	30	Bov non-clin	Cerro	6/9/08	PAN0	Y	CU-213
740	LR-1220-08	FSL R8-1749	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
741	LR-1220-09	FSL R8-1750	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
742	LR-1220-10	FSL R8-1751	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
743	LR-1220-11	FSL R8-1752	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
744	LR-1220-12	FSL R8-1753	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
745	LR-1220-13	FSL R8-1754	30	Bov non-clin	Cerro	6/9/08	PAN0	N	
534	LR-1183-01	FSL R8-1407	30	Environmental	Cerro	6/16/08	PAN0	Y	CU-213
815	LR-1248-01	FSL R8-2059	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
816	LR-1248-02	FSL R8-2060	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
817	LR-1248-03	FSL R8-2061	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
818	LR-1248-04	FSL R8-2062	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
819	LR-1248-05	FSL R8-2063	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
820	LR-1248-06	FSL R8-2064	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
821	LR-1248-07	FSL R8-2065	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
822	LR-1248-08	FSL R8-2066	30	Bov non-clin	Cerro	6/23/08	PAN0	Y	CU-213
823	LR-1248-09	FSL R8-2067	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
824	LR-1248-10	FSL R8-2068	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
825	LR-1248-11	FSL R8-2069	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
826	LR-1248-12	FSL R8-2070	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
827	LR-1248-13	FSL R8-2071	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
828	LR-1248-14	FSL R8-2072	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
829	LR-1248-15	FSL R8-2073	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
830	LR-1248-16	FSL R8-2074	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
831	LR-1248-17	FSL R8-2075	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
832	LR-1248-18	FSL R8-2076	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
833	LR-1248-19	FSL R8-2077	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
834	LR-1248-20	FSL R8-2078	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
835	LR-1248-21	FSL R8-2079	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
836	LR-1248-22	FSL R8-2080	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
837	LR-1248-23	FSL R8-2081	30	Bov non-clin	Cerro	6/23/08	PAN0	N	
876	LR-1260-01	FSL R8-2252	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
877	LR-1260-02	FSL R8-2253	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
878	LR-1260-03	FSL R8-2254	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
879	LR-1260-04	FSL R8-2255	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
880	LR-1260-05	FSL R8-2256	30	Bov non-clin	Cerro	7/7/08	PAN0	N	

Appendix Table 1

881	LR-1260-06	FSL R8-2257	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
882	LR-1260-07	FSL R8-2258	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
883	LR-1260-08	FSL R8-2259	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
884	LR-1260-09	FSL R8-2260	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
885	LR-1260-10	FSL R8-2261	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
886	LR-1260-11	FSL R8-2262	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
887	LR-1260-12	FSL R8-2263	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
888	LR-1260-13	FSL R8-2264	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
889	LR-1260-14	FSL R8-2265	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
890	LR-1260-15	FSL R8-2266	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
891	LR-1260-16	FSL R8-2267	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
892	LR-1260-17	FSL R8-2268	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
893	LR-1260-18	FSL R8-2269	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
894	LR-1260-19	FSL R8-2270	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
895	LR-1260-20	FSL R8-2271	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
896	LR-1260-21	FSL R8-2272	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
897	LR-1260-22	FSL R8-2273	30	Bov non-clin	Cerro	7/7/08	PAN0	Y	CU-213
898	LR-1260-23	FSL R8-2274	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
899	LR-1260-24	FSL R8-2275	30	Bov non-clin	Cerro	7/7/08	PAN0	N	
622	LR-1202-01	FSL R8-1630	30	Environmental	Cerro	7/21/08	PAN0	Y	CU-213
935	LR-1266-01	FSL R8-2399	30	Bov non-clin	Cerro	7/21/08	PAN0	N	
936	LR-1266-02	FSL R8-2400	30	Bov non-clin	Cerro	7/21/08	PAN0	N	
937	LR-1266-03	FSL R8-2401	30	Bov non-clin	Cerro	7/21/08	PAN0	Y	CU-213
938	LR-1266-04	FSL R8-2402	30	Bov non-clin	Cerro	7/21/08	PAN0	N	
939	LR-1266-05	FSL R8-2403	30	Bov non-clin	Cerro	7/21/08	PAN0	N	
948	LR-1269-01	FSL R8-2412	30	Bov non-clin	Cerro	8/4/08	PAN0	Y	CU-213
949	LR-1269-02	FSL R8-2413	30	Bov non-clin	Cerro	8/4/08	PAN0	N	
950	LR-1269-03	FSL R8-2414	30	Bov non-clin	Cerro	8/4/08	PAN0	N	
951	LR-1269-04	FSL R8-2415	30	Bov non-clin	Cerro	8/4/08	PAN0	N	
952	LR-1269-05	FSL R8-2416	30	Bov non-clin	Cerro	8/4/08	PAN0	N	
953	LR-1269-06	FSL R8-2417	30	Bov non-clin	Cerro	8/4/08	PAN0	N	
954	LR-1269-07	FSL R8-2418	30	Bov non-clin	Cerro	8/4/08	PAN0	N	
775	LR-1234-01	FSL R8-2019	30	Environmental	Cerro	9/4/08	PAN0	Y	CU-213
776	LR-1234-02	FSL R8-2020	30	Environmental	Cerro	9/4/08	PAN0	N	
777	LR-1234-03	FSL R8-2021	30	Environmental	Cerro	9/4/08	PAN0	N	
1069	LR-1291-01	FSL R8-2819	30	Environmental	Cerro	10/15/08	PAN0	N	
1070	LR-1291-02	FSL R8-2820	30	Environmental	Cerro	10/15/08	PAN0	N	
1071	LR-1291-03	FSL R8-2821	30	Environmental	Cerro	10/15/08	PAN0	Y	CU-213
628	LR-1206-01	FSL R8-1637	35	Environmental	Newport	7/23/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
629	LR-1206-02	FSL R8-1638	35	Environmental	Newport	7/23/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-126
630	LR-1206-03	FSL R8-1639	35	Environmental	Newport	7/23/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
872	LR-1259-01	FSL R8-2248	35	Environmental	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
873	LR-1259-02	FSL R8-2249	35	Environmental	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-1032
874	LR-1259-03	FSL R8-2250	35	Environmental	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
875	LR-1259-04	FSL R8-2251	35	Environmental	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
999	LR-1278-01	FSL R8-2678	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1000	LR-1278-02	FSL R8-2679	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1001	LR-1278-03	FSL R8-2680	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1002	LR-1278-04	FSL R8-2681	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1003	LR-1278-05	FSL R8-2682	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1004	LR-1278-06	FSL R8-2683	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1005	LR-1278-07	FSL R8-2684	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1006	LR-1278-08	FSL R8-2685	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-126
1007	LR-1278-09	FSL R8-2686	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1008	LR-1278-10	FSL R8-2687	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1009	LR-1279-01	FSL R8-2688	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 STR1 SUL1 TEL1	Y	CU-126
1010	LR-1278-11	FSL R8-2689	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	

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1011	LR-1280-01	FSL R8-2690	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CHL1 STR1 SUL1 TEL1	Y	CU-126
1012	LR-1278-12	FSL R8-2691	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1013	LR-1278-13	FSL R8-2692	35	Bov non-clin	Newport	10/2/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1093	LR-1304-01	FSL R8-2843	35	Environmental	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-126
1094	LR-1304-02	FSL R8-2844	35	Environmental	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1095	LR-1304-03	FSL R8-2845	35	Environmental	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1143	LR-1324-01	FSL R8-2893	35	Bov non-clin	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1144	LR-1324-02	FSL R8-2894	35	Bov non-clin	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-126
1145	LR-1324-03	FSL R8-2895	35	Bov non-clin	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1146	LR-1324-04	FSL R8-2896	35	Bov non-clin	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1147	LR-1324-05	FSL R8-2897	35	Bov non-clin	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1148	LR-1324-06	FSL R8-2898	35	Bov non-clin	Newport	11/5/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
1164	LR-1328-01	FSL R8-3136	35	Environmental	Newport	12/9/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-126
1218	LR-1344-01	FSL R8-3190	35	Bov non-clin	Newport	12/9/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-126
1219	LR-1344-02	FSL R8-3191	35	Bov non-clin	Newport	12/9/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	N	
43	LR-1028-01	FSL R6-931	36	Environmental	Infantis	11/13/07	PAN0	N	
44	LR-1028-02	FSL R6-932	36	Environmental	Infantis	11/13/07	PAN0	Y	CU-114
39	LR-1026-01	FSL R6-927	39	Environmental	Anatum	11/13/07	PAN0	N	
40	LR-1026-02	FSL R6-928	39	Environmental	Anatum	11/13/07	PAN0	Y	CU-424
41	LR-1027-01	FSL R6-929	39	Environmental	Anatum var. 15+	11/13/07	PAN0	Y	CU-423
42	LR-1027-02	FSL R6-930	39	Environmental	Anatum var. 15+	11/13/07	PAN0	N	
192	LR-1083-01	FSL R8-187	39	Environmental	Anatum	1/9/08	PAN0	Y	CU-424
193	LR-1083-02	FSL R8-188	39	Environmental	Anatum	1/9/08	PAN0	N	
266	LR-1105-01	FSL R8-261	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
267	LR-1105-02	FSL R8-262	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
268	LR-1105-03	FSL R8-263	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
269	LR-1105-04	FSL R8-264	39	Bov non-clin	Anatum	1/9/08	PAN0	Y	CU-1025
270	LR-1105-05	FSL R8-265	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
271	LR-1105-06	FSL R8-266	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
272	LR-1105-07	FSL R8-267	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
273	LR-1105-08	FSL R8-268	39	Bov non-clin	Anatum	1/9/08	PAN0	N	
108	LR-1056-01	FSL R8-347	39	Environmental	Anatum var. 15+	2/21/08	PAN0	N	
109	LR-1056-02	FSL R8-348	39	Environmental	Anatum var. 15+	2/21/08	PAN0	Y	CU-423
308	LR-1124-01	FSL R8-841	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
309	LR-1125-01	FSL R8-842	39	Bov non-clin	Anatum	2/21/08	PAN0	Y	CU-424
310	LR-1124-02	FSL R8-843	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
311	LR-1124-03	FSL R8-844	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
312	LR-1124-04	FSL R8-845	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
313	LR-1124-05	FSL R8-846	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
314	LR-1126-01	FSL R8-847	39	Bov non-clin	Anatum var. 15+	2/21/08	AMP1	Y	CU-423
315	LR-1124-06	FSL R8-848	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
316	LR-1124-07	FSL R8-849	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
317	LR-1124-08	FSL R8-850	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
318	LR-1124-09	FSL R8-851	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	Y	CU-423
319	LR-1124-10	FSL R8-852	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
320	LR-1124-11	FSL R8-853	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
321	LR-1124-12	FSL R8-854	39	Bov non-clin	Anatum var. 15+	2/21/08	PAN0	N	
355	LR-1143-01	FSL R8-905	39	Environmental	Anatum	4/17/08	PAN0	Y	CU-424
356	LR-1143-02	FSL R8-906	39	Environmental	Anatum	4/17/08	PAN0	N	
1110	LR-1315-01	FSL R8-2860	39	Environmental	Anatum* (Untypeable)	12/3/08	PAN0	Y	CU-424
45	LR-1029-01	FSL R6-933	40	Environmental	Mbandaka	11/14/07	PAN0	Y	CU-758
46	LR-1029-02	FSL R6-934	40	Environmental	Mbandaka	11/14/07	PAN0	N	
194	LR-1084-01	FSL R8-189	40	Environmental	Mbandaka	1/10/08	PAN0	Y	CU-758
195	LR-1084-02	FSL R8-190	40	Environmental	Mbandaka	1/10/08	PAN0	N	
761	LR-1222-01	FSL R8-2004	41	Environmental	Montevideo	8/4/08	PAN0	Y	CU-156
762	LR-1222-02	FSL R8-2005	41	Environmental	Montevideo	8/4/08	PAN0	N	
1062	LR-1288-01	FSL R8-2812	41	Environmental	Montevideo	10/3/08	PAN0	Y	CU-156
1118	LR-1319-01	FSL R8-2868	41	Bov non-clin	Montevideo	10/3/08	PAN0	Y	CU-156

Appendix Table 1

47	LR-1030-01	FSL R6-935	42	Environmental	Kentucky	11/15/07	PAN0	Y	CU-96
196	LR-1085-01	FSL R8-191	42	Environmental	Kentucky	1/17/08	PAN0	Y	CU-96
274	LR-1106-01	FSL R8-269	42	Bov non-clin	Kentucky	1/17/08	PAN0	Y	CU-96
275	LR-1106-02	FSL R8-270	42	Bov non-clin	Kentucky	1/17/08	PAN0	N	
330	LR-1129-01	FSL R8-863	42	Bov non-clin	Kentucky	3/13/08	PAN0	Y	CU-96
331	LR-1129-02	FSL R8-864	42	Bov non-clin	Kentucky	3/13/08	PAN0	N	
363	LR-1147-01	FSL R8-913	42	Environmental	Kentucky	4/23/08	AMC1 AMP1 CHL2 TEL1	Y	CU-96
364	LR-1148-01	FSL R8-914	42	Environmental	Kentucky	4/23/08	PAN0	Y	CU-96
421	LR-1162-01	FSL R8-971	42	Bov non-clin	Kentucky	4/23/08	PAN0	Y	CU-96
631	LR-1207-01	FSL R8-1640	42	Environmental	Kentucky	7/23/08	AMC1 AMP1 CHL2 TEL1	Y	CU-96
766	LR-1226-01	FSL R8-2009	42	Environmental	Kentucky	8/19/08	PAN0	Y	CU-96
860	LR-1251-01	FSL R8-2236	42	Environmental	Kentucky* (Untypeable)	9/17/08	AMC1 AMP1 TEL1	Y	CU-96
137	LR-1061-01	FSL R8-059	45	Environmental	Cerro	11/29/07	PAN0	Y	CU-213
138	LR-1062-01	FSL R8-060	46	Environmental	Agona	11/29/07	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1 SXT1	Y	CU-807
288	LR-1114-01	FSL R8-821	46	Bov non-clin	Senftenberg	1/29/08	PAN0	Y	CU-372
864	LR-1253-01	FSL R8-2240	46	Environmental	Tennessee	9/25/08	PAN0	Y	CU-442
865	LR-1254-01	FSL R8-2241	46	Environmental	Heidelberg	9/25/08	AMC1 AMP1 FOX1 CEF1 CRO2 KAN1 STR1 TEL1	Y	CU-1016
790	LR-1243-01	FSL R8-2034	47	Environmental	Cerro	9/11/08	PAN0	Y	CU-213
791	LR-1244-01	FSL R8-2035	47	Environmental	3,10:-:1,5* (Not serotyped)	9/11/08	PAN0	Y	CU-007
1077	LR-1295-01	FSL R8-2827	47	Environmental	Cerro	10/23/08	PAN0	Y	CU-213
1078	LR-1296-01	FSL R8-2828	47	Environmental	3,10:-:1,5	10/23/08	PAN0	Y	CU-007
1172	LR-1333-01	FSL R8-3144	47	Environmental	3,10:-:1,5	1/6/09	PAN0	N	
1173	LR-1333-02	FSL R8-3145	47	Environmental	3,10:-:1,5	1/6/09	PAN0	Y	CU-007
1174	LR-1334-01	FSL R8-3146	47	Environmental	Cerro	1/6/09	PAN0	Y	CU-213
1220	LR-1345-01	FSL R8-3192	47	Bov non-clin	Cerro	1/6/09	PAN0	Y	CU-213
1221	LR-1346-01	FSL R8-3193	47	Bov non-clin	3,10:-:1,5	1/6/09	KAN1 TEL2	Y	CU-007
1222	LR-1347-01	FSL R8-3194	47	Bov non-clin	3,10:-:1,5* (Lexington)	1/6/09	PAN0	Y	CU-007
113	LR-1058-01	FSL R8-352	48	Environmental	Mbandaka	Not given	PAN0	Y	CU-758
97	LR-1050-01	FSL R8-336	49	Environmental	Cerro	1/31/08	PAN0	N	
98	LR-1050-02	FSL R8-337	49	Environmental	Cerro	1/31/08	PAN0	Y	CU-213
99	LR-1050-03	FSL R8-338	49	Environmental	Cerro	1/31/08	PAN0	N	
116	LR-1060-01	FSL R8-355	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
117	LR-1060-02	FSL R8-356	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
118	LR-1060-03	FSL R8-357	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
119	LR-1060-04	FSL R8-358	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
120	LR-1060-05	FSL R8-359	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
121	LR-1060-06	FSL R8-360	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
122	LR-1060-07	FSL R8-361	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
123	LR-1060-08	FSL R8-362	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
124	LR-1060-09	FSL R8-363	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
125	LR-1060-10	FSL R8-364	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
126	LR-1060-11	FSL R8-365	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
127	LR-1060-12	FSL R8-366	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
128	LR-1060-13	FSL R8-367	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
129	LR-1060-14	FSL R8-368	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
130	LR-1060-15	FSL R8-369	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
131	LR-1060-16	FSL R8-370	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
132	LR-1060-17	FSL R8-371	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
133	LR-1060-18	FSL R8-372	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
134	LR-1060-19	FSL R8-373	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
135	LR-1060-20	FSL R8-374	49	Bov non-clin	Cerro	1/31/08	PAN0	N	
136	LR-1060-21	FSL R8-375	49	Bov non-clin	Cerro	1/31/08	PAN0	Y	CU-213
343	LR-1136-01	FSL R8-893	49	Environmental	Cerro	4/1/08	PAN0	Y	CU-213
344	LR-1136-02	FSL R8-894	49	Environmental	Cerro	4/1/08	PAN0	N	
345	LR-1136-03	FSL R8-895	49	Environmental	Cerro	4/1/08	PAN0	N	
391	LR-1158-01	FSL R8-941	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
392	LR-1158-02	FSL R8-942	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
393	LR-1158-03	FSL R8-943	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
394	LR-1158-04	FSL R8-944	49	Bov non-clin	Cerro	4/1/08	PAN0	N	

395	LR-1158-05	FSL R8-945	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
396	LR-1158-06	FSL R8-946	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
397	LR-1158-07	FSL R8-947	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
398	LR-1158-08	FSL R8-948	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
399	LR-1158-09	FSL R8-949	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
400	LR-1158-10	FSL R8-950	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
401	LR-1158-11	FSL R8-951	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
402	LR-1158-12	FSL R8-952	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
403	LR-1158-13	FSL R8-953	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
404	LR-1158-14	FSL R8-954	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
405	LR-1158-15	FSL R8-955	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
406	LR-1158-16	FSL R8-956	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
407	LR-1158-17	FSL R8-957	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
408	LR-1158-18	FSL R8-958	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
409	LR-1158-19	FSL R8-959	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
410	LR-1158-20	FSL R8-960	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
411	LR-1158-21	FSL R8-961	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
412	LR-1158-22	FSL R8-962	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
413	LR-1158-23	FSL R8-963	49	Bov non-clin	Cerro	4/1/08	PAN0	Y	CU-213
414	LR-1158-24	FSL R8-964	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
415	LR-1158-25	FSL R8-965	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
416	LR-1158-26	FSL R8-966	49	Bov non-clin	Cerro	4/1/08	PAN0	N	
515	LR-1172-01	FSL R8-1388	49	Environmental	Cerro	5/6/08	PAN0	N	
516	LR-1172-02	FSL R8-1389	49	Environmental	Cerro	5/6/08	PAN0	N	
517	LR-1172-03	FSL R8-1390	49	Environmental	Cerro	5/6/08	PAN0	Y	CU-840
518	LR-1172-04	FSL R8-1391	49	Environmental	Cerro	5/6/08	PAN0	N	
548	LR-1191-01	FSL R8-1421	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
549	LR-1191-02	FSL R8-1422	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
550	LR-1191-03	FSL R8-1423	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
551	LR-1191-04	FSL R8-1424	49	Bov non-clin	Cerro	5/6/08	PAN0	Y	CU-213
552	LR-1191-05	FSL R8-1425	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
553	LR-1191-06	FSL R8-1426	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
554	LR-1191-07	FSL R8-1427	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
555	LR-1191-08	FSL R8-1428	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
556	LR-1191-09	FSL R8-1429	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
557	LR-1191-10	FSL R8-1430	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
558	LR-1191-11	FSL R8-1431	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
559	LR-1191-12	FSL R8-1432	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
560	LR-1191-13	FSL R8-1433	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
561	LR-1191-14	FSL R8-1434	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
562	LR-1191-15	FSL R8-1435	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
563	LR-1191-16	FSL R8-1436	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
564	LR-1191-17	FSL R8-1437	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
565	LR-1191-18	FSL R8-1438	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
566	LR-1191-19	FSL R8-1439	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
567	LR-1191-20	FSL R8-1440	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
568	LR-1191-21	FSL R8-1441	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
569	LR-1192-01	FSL R8-1442	49	Bov non-clin	Cerro	5/6/08	AMP1	Y	CU-213
570	LR-1191-22	FSL R8-1443	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
571	LR-1191-23	FSL R8-1444	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
572	LR-1191-24	FSL R8-1445	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
573	LR-1191-25	FSL R8-1446	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
574	LR-1191-26	FSL R8-1447	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
575	LR-1191-27	FSL R8-1448	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
576	LR-1191-28	FSL R8-1449	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
577	LR-1191-29	FSL R8-1450	49	Bov non-clin	Cerro	5/6/08	PAN0	N	

Appendix Table 1

578	LR-1191-30	FSL R8-1451	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
579	LR-1191-31	FSL R8-1452	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
580	LR-1191-32	FSL R8-1453	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
581	LR-1191-33	FSL R8-1454	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
582	LR-1193-01	FSL R8-1455	49	Bov non-clin	Cerro	5/6/08	TEL1	Y	CU-213
583	LR-1191-34	FSL R8-1456	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
584	LR-1191-35	FSL R8-1457	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
585	LR-1191-36	FSL R8-1458	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
586	LR-1191-37	FSL R8-1459	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
587	LR-1191-38	FSL R8-1460	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
588	LR-1191-39	FSL R8-1461	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
589	LR-1191-40	FSL R8-1462	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
590	LR-1191-41	FSL R8-1463	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
591	LR-1191-42	FSL R8-1464	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
592	LR-1191-43	FSL R8-1465	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
593	LR-1191-44	FSL R8-1466	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
594	LR-1191-45	FSL R8-1467	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
595	LR-1191-46	FSL R8-1468	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
596	LR-1191-47	FSL R8-1469	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
597	LR-1191-48	FSL R8-1470	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
598	LR-1191-49	FSL R8-1471	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
599	LR-1191-50	FSL R8-1472	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
600	LR-1191-51	FSL R8-1473	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
601	LR-1191-52	FSL R8-1474	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
602	LR-1191-53	FSL R8-1475	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
603	LR-1191-54	FSL R8-1476	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
604	LR-1191-55	FSL R8-1477	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
605	LR-1191-56	FSL R8-1478	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
606	LR-1191-57	FSL R8-1479	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
607	LR-1191-58	FSL R8-1480	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
608	LR-1191-59	FSL R8-1481	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
609	LR-1191-60	FSL R8-1482	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
610	LR-1191-61	FSL R8-1483	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
611	LR-1191-62	FSL R8-1484	49	Bov non-clin	Cerro	5/6/08	PAN0	N	
717	LR-1219-01	FSL R8-1726	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
718	LR-1219-02	FSL R8-1727	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
719	LR-1219-03	FSL R8-1728	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
720	LR-1219-04	FSL R8-1729	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
721	LR-1219-05	FSL R8-1730	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
722	LR-1219-06	FSL R8-1731	49	Bov non-clin	Cerro	6/2/08	PAN0	Y	CU-213
723	LR-1219-07	FSL R8-1732	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
724	LR-1219-08	FSL R8-1733	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
725	LR-1219-09	FSL R8-1734	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
726	LR-1219-10	FSL R8-1735	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
727	LR-1219-11	FSL R8-1736	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
728	LR-1219-12	FSL R8-1737	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
729	LR-1219-13	FSL R8-1738	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
730	LR-1219-14	FSL R8-1739	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
731	LR-1219-15	FSL R8-1740	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
732	LR-1219-16	FSL R8-1741	49	Bov non-clin	Cerro	6/2/08	PAN0	N	
524	LR-1176-01	FSL R8-1397	49	Environmental	Cerro	6/9/08	PAN0	Y	CU-213
525	LR-1176-02	FSL R8-1398	49	Environmental	Cerro	6/9/08	PAN0	N	
526	LR-1176-03	FSL R8-1399	49	Environmental	Cerro	6/9/08	PAN0	N	
746	LR-1221-01	FSL R8-1755	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
747	LR-1221-02	FSL R8-1756	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
748	LR-1221-03	FSL R8-1757	49	Bov non-clin	Cerro	6/16/08	PAN0	N	

Appendix Table 1

749	LR-1221-04	FSL R8-1758	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
750	LR-1221-05	FSL R8-1759	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
751	LR-1221-06	FSL R8-1760	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
752	LR-1221-07	FSL R8-1761	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
753	LR-1221-08	FSL R8-1762	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
754	LR-1221-09	FSL R8-1763	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
755	LR-1221-10	FSL R8-1764	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
756	LR-1221-11	FSL R8-1765	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
757	LR-1221-12	FSL R8-1766	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
758	LR-1221-13	FSL R8-1767	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
759	LR-1221-14	FSL R8-1768	49	Bov non-clin	Cerro	6/16/08	PAN0	N	
760	LR-1221-15	FSL R8-1769	49	Bov non-clin	Cerro	6/16/08	PAN0	Y	CU-213
838	LR-1249-01	FSL R8-2082	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
839	LR-1249-02	FSL R8-2083	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
840	LR-1249-03	FSL R8-2084	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
841	LR-1249-04	FSL R8-2085	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
842	LR-1249-05	FSL R8-2086	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
843	LR-1249-06	FSL R8-2087	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
844	LR-1249-07	FSL R8-2088	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
845	LR-1249-08	FSL R8-2089	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
846	LR-1249-09	FSL R8-2090	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
847	LR-1249-10	FSL R8-2091	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
848	LR-1249-11	FSL R8-2092	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
849	LR-1249-12	FSL R8-2093	49	Bov non-clin	Cerro	6/30/08	PAN0	Y	CU-213
850	LR-1249-13	FSL R8-2094	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
851	LR-1249-14	FSL R8-2095	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
852	LR-1249-15	FSL R8-2096	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
853	LR-1249-16	FSL R8-2097	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
854	LR-1249-17	FSL R8-2098	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
855	LR-1249-18	FSL R8-2099	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
856	LR-1249-19	FSL R8-2100	49	Bov non-clin	Cerro	6/30/08	PAN0	N	
925	LR-1265-01	FSL R8-2389	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
926	LR-1265-02	FSL R8-2390	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
927	LR-1265-03	FSL R8-2391	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
928	LR-1265-04	FSL R8-2392	49	Bov non-clin	Cerro	7/14/08	PAN0	Y	CU-213
929	LR-1265-05	FSL R8-2393	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
930	LR-1265-06	FSL R8-2394	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
931	LR-1265-07	FSL R8-2395	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
932	LR-1265-08	FSL R8-2396	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
933	LR-1265-09	FSL R8-2397	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
934	LR-1265-10	FSL R8-2398	49	Bov non-clin	Cerro	7/14/08	PAN0	N	
940	LR-1267-01	FSL R8-2404	49	Bov non-clin	Cerro	7/28/08	PAN0	N	
941	LR-1267-02	FSL R8-2405	49	Bov non-clin	Cerro	7/28/08	PAN0	N	
942	LR-1267-03	FSL R8-2406	49	Bov non-clin	Cerro	7/28/08	PAN0	N	
943	LR-1267-04	FSL R8-2407	49	Bov non-clin	Cerro	7/28/08	PAN0	N	
944	LR-1267-05	FSL R8-2408	49	Bov non-clin	Cerro	7/28/08	PAN0	N	
945	LR-1267-06	FSL R8-2409	49	Bov non-clin	Cerro	7/28/08	PAN0	Y	CU-213
946	LR-1268-01	FSL R8-2410	49	Bov non-clin	Minnesota	7/28/08	CHL2	Y	CU-1029
947	LR-1267-07	FSL R8-2411	49	Bov non-clin	Cerro	7/28/08	PAN0	N	
1014	LR-1281-01	FSL R8-2693	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1015	LR-1282-01	FSL R8-2694	49	Bov non-clin	Minnesota	8/11/08	PAN0	Y	CU-1029
1016	LR-1281-02	FSL R8-2695	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1017	LR-1281-03	FSL R8-2696	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1018	LR-1281-04	FSL R8-2697	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1019	LR-1281-05	FSL R8-2698	49	Bov non-clin	Cerro	8/11/08	PAN0	Y	CU-213
1020	LR-1281-06	FSL R8-2699	49	Bov non-clin	Cerro	8/11/08	PAN0	N	

Appendix Table 1

1021	LR-1281-07	FSL R8-2700	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1022	LR-1281-08	FSL R8-2701	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1023	LR-1281-09	FSL R8-2702	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1024	LR-1281-10	FSL R8-2703	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1025	LR-1281-11	FSL R8-2704	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1026	LR-1281-12	FSL R8-2705	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1027	LR-1281-13	FSL R8-2706	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1028	LR-1281-14	FSL R8-2707	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1029	LR-1281-15	FSL R8-2708	49	Bov non-clin	Cerro	8/11/08	PAN0	N	
1030	LR-1283-01	FSL R8-2709	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1031	LR-1283-02	FSL R8-2710	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1032	LR-1283-03	FSL R8-2711	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1033	LR-1283-04	FSL R8-2712	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1034	LR-1283-05	FSL R8-2713	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1035	LR-1283-06	FSL R8-2714	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1036	LR-1283-07	FSL R8-2715	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1037	LR-1283-08	FSL R8-2716	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1038	LR-1283-09	FSL R8-2717	49	Bov non-clin	Cerro	8/25/08	PAN0	Y	CU-213
1039	LR-1283-10	FSL R8-2718	49	Bov non-clin	Cerro	8/25/08	PAN0	N	
1040	LR-1284-01	FSL R8-2719	49	Bov non-clin	Cerro	9/8/08	PAN0	Y	CU-213
1041	LR-1284-02	FSL R8-2720	49	Bov non-clin	Cerro	9/8/08	PAN0	N	
1042	LR-1284-03	FSL R8-2721	49	Bov non-clin	Cerro	9/8/08	PAN0	N	
1043	LR-1284-04	FSL R8-2722	49	Bov non-clin	Cerro	9/8/08	PAN0	N	
1044	LR-1284-05	FSL R8-2723	49	Bov non-clin	Cerro	9/8/08	PAN0	N	
1045	LR-1284-06	FSL R8-2724	49	Bov non-clin	Cerro	9/8/08	PAN0	N	
1046	LR-1284-07	FSL R8-2725	49	Bov non-clin	Cerro	9/8/08	PAN0	N	
861	LR-1252-01	FSL R8-2237	49	Environmental	Cerro	9/22/08	PAN0	Y	CU-839
862	LR-1252-02	FSL R8-2238	49	Environmental	Cerro	9/22/08	PAN0	N	
863	LR-1252-03	FSL R8-2239	49	Environmental	Cerro	9/22/08	PAN0	N	
1047	LR-1285-01	FSL R8-2726	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1048	LR-1285-02	FSL R8-2727	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1049	LR-1285-03	FSL R8-2728	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1050	LR-1285-04	FSL R8-2729	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1051	LR-1285-05	FSL R8-2730	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1052	LR-1285-06	FSL R8-2731	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1053	LR-1285-07	FSL R8-2732	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1054	LR-1285-08	FSL R8-2733	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1055	LR-1285-09	FSL R8-2734	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1056	LR-1285-10	FSL R8-2735	49	Bov non-clin	Cerro	9/22/08	PAN0	Y	CU-213
1057	LR-1285-11	FSL R8-2736	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1058	LR-1285-12	FSL R8-2737	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1059	LR-1285-13	FSL R8-2738	49	Bov non-clin	Cerro	9/22/08	PAN0	N	
1060	LR-1286-01	FSL R8-2739	49	Bov non-clin	Cerro	10/6/08	PAN0	Y	CU-213
1182	LR-1338-01	FSL R8-3154	49	Environmental	Cerro	1/15/09	PAN0	N	
1183	LR-1338-02	FSL R8-3155	49	Environmental	Cerro	1/15/09	PAN0	Y	CU-973
1184	LR-1338-03	FSL R8-3156	49	Environmental	Cerro	1/15/09	PAN0	N	
1277	LR-1361-01	FSL R8-3435	49	Environmental	Cerro	2/23/09	PAN0	N	
1278	LR-1361-02	FSL R8-3436	49	Environmental	Cerro	2/23/09	PAN0	N	
1279	LR-1361-03	FSL R8-3437	49	Environmental	Cerro	2/23/09	PAN0	Y	CU-213
1280	LR-1361-04	FSL R8-3438	49	Environmental	Cerro	2/23/09	PAN0	N	
1090	LR-1303-01	FSL R8-2840	50	Environmental	Cerro	11/1/08	PAN0	N	
1091	LR-1303-02	FSL R8-2841	50	Environmental	Cerro	11/1/08	PAN0	N	
1092	LR-1303-03	FSL R8-2842	50	Environmental	Cerro	11/1/08	PAN0	Y	CU-213
1107	LR-1314-01	FSL R8-2857	50	Environmental	Cerro	12/3/08	PAN0	Y	CU-213
1108	LR-1314-02	FSL R8-2858	50	Environmental	Cerro	12/3/08	PAN0	N	
1109	LR-1314-03	FSL R8-2859	50	Environmental	Cerro	12/3/08	PAN0	N	
1188	LR-1341-01	FSL R8-3160	50	Bov non-clin	Cerro	12/3/08	PAN0	N	

Appendix Table 1

1189	LR-1341-02	FSL R8-3161	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1190	LR-1341-03	FSL R8-3162	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1191	LR-1341-04	FSL R8-3163	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1192	LR-1341-05	FSL R8-3164	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1193	LR-1341-06	FSL R8-3165	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1194	LR-1341-07	FSL R8-3166	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1195	LR-1341-08	FSL R8-3167	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1196	LR-1341-09	FSL R8-3168	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1197	LR-1341-10	FSL R8-3169	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1198	LR-1341-11	FSL R8-3170	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1199	LR-1341-12	FSL R8-3171	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1200	LR-1341-13	FSL R8-3172	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1201	LR-1341-14	FSL R8-3173	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1202	LR-1341-15	FSL R8-3174	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1203	LR-1341-16	FSL R8-3175	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1204	LR-1341-17	FSL R8-3176	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1205	LR-1341-18	FSL R8-3177	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1206	LR-1341-19	FSL R8-3178	50	Bov non-clin	Cerro	12/3/08	PAN0	Y	CU-213
1207	LR-1341-20	FSL R8-3179	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1208	LR-1341-21	FSL R8-3180	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1209	LR-1341-22	FSL R8-3181	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1210	LR-1341-23	FSL R8-3182	50	Bov non-clin	Cerro	12/3/08	PAN0	N	
1178	LR-1336-01	FSL R8-3150	50	Environmental	Cerro	1/9/09	PAN0	Y	CU-213
1179	LR-1336-02	FSL R8-3151	50	Environmental	Cerro	1/9/09	PAN0	N	
1223	LR-1348-01	FSL R8-3195	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1224	LR-1348-02	FSL R8-3196	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1225	LR-1348-03	FSL R8-3197	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1226	LR-1348-04	FSL R8-3198	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1227	LR-1348-05	FSL R8-3199	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1228	LR-1348-06	FSL R8-3200	50	Bov non-clin	Cerro	1/9/09	PAN0	Y	CU-213
1229	LR-1348-07	FSL R8-3201	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1230	LR-1348-08	FSL R8-3202	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1231	LR-1348-09	FSL R8-3203	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1232	LR-1348-10	FSL R8-3204	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1233	LR-1348-11	FSL R8-3205	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1234	LR-1348-12	FSL R8-3206	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1235	LR-1348-13	FSL R8-3207	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1236	LR-1348-14	FSL R8-3208	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1237	LR-1348-15	FSL R8-3209	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1238	LR-1348-16	FSL R8-3210	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1239	LR-1348-17	FSL R8-3211	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1240	LR-1348-18	FSL R8-3212	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1241	LR-1348-19	FSL R8-3213	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1242	LR-1348-20	FSL R8-3214	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1243	LR-1348-21	FSL R8-3215	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1244	LR-1348-22	FSL R8-3216	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1245	LR-1348-23	FSL R8-3217	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1246	LR-1348-24	FSL R8-3218	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1247	LR-1348-25	FSL R8-3219	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1248	LR-1348-26	FSL R8-3220	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1249	LR-1348-27	FSL R8-3221	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1250	LR-1348-28	FSL R8-3222	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1251	LR-1348-29	FSL R8-3223	50	Bov non-clin	Cerro	1/9/09	PAN0	N	
1264	LR-1356-01	FSL R8-3422	50	Environmental	Cerro	2/9/09	PAN0	Y	CU-213
1265	LR-1356-02	FSL R8-3423	50	Environmental	Cerro	2/9/09	PAN0	N	
1266	LR-1356-03	FSL R8-3424	50	Environmental	Cerro	2/9/09	PAN0	N	

Appendix Table 1

1302	LR-1368-01	FSL R8-3460	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1303	LR-1368-02	FSL R8-3461	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1304	LR-1368-03	FSL R8-3462	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1305	LR-1368-04	FSL R8-3463	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1306	LR-1368-05	FSL R8-3464	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1307	LR-1368-06	FSL R8-3465	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1308	LR-1368-07	FSL R8-3466	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1309	LR-1368-08	FSL R8-3467	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1310	LR-1368-09	FSL R8-3468	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1311	LR-1368-10	FSL R8-3469	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1312	LR-1368-11	FSL R8-3470	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1313	LR-1368-12	FSL R8-3471	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1314	LR-1368-13	FSL R8-3472	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1315	LR-1368-14	FSL R8-3473	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1316	LR-1368-15	FSL R8-3474	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1317	LR-1368-16	FSL R8-3475	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1318	LR-1368-17	FSL R8-3476	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1319	LR-1368-18	FSL R8-3477	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1320	LR-1368-19	FSL R8-3478	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1321	LR-1368-20	FSL R8-3479	50	Bov non-clin	Cerro	2/9/09	PAN0	Y	CU-213
1322	LR-1368-21	FSL R8-3480	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1323	LR-1368-22	FSL R8-3481	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1324	LR-1368-23	FSL R8-3482	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1325	LR-1368-24	FSL R8-3483	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
1326	LR-1368-25	FSL R8-3484	50	Bov non-clin	Cerro	2/9/09	PAN0	N	
370	LR-1153-01	FSL R8-920	51	Environmental	Typhimurium	5/9/08	PAN0	Y	CU-723
365	LR-1149-01	FSL R8-915	52	Environmental	4,5,12:i:-* (Kentucky)	4/26/08	AMC1 AMP1 FOX1 CEF1 CRO2	Y	CU-1024
527	LR-1177-01	FSL R8-1400	52	Environmental	Anatum	6/11/08	PAN0	Y	CU-430
763	LR-1223-01	FSL R8-2006	52	Environmental	Mbandaka	8/19/08	PAN0	Y	CU-758
764	LR-1224-01	FSL R8-2007	52	Environmental	Mbandaka (Cerro)	8/19/08	AMC2 AMP1 FOX1 CEF1 STR1 TEL1	Y	CU-758
765	LR-1225-01	FSL R8-2008	52	Environmental	Cerro	8/19/08	PAN0	Y	CU-843
956	LR-1271-01	FSL R8-2635	52	Bov non-clin	Cerro	8/19/08	PAN0	N	
957	LR-1271-02	FSL R8-2636	52	Bov non-clin	Cerro	8/19/08	PAN0	N	
958	LR-1271-03	FSL R8-2637	52	Bov non-clin	Cerro	8/19/08	PAN0	N	
959	LR-1271-04	FSL R8-2638	52	Bov non-clin	Cerro	8/19/08	PAN0	N	
960	LR-1271-05	FSL R8-2639	52	Bov non-clin	Cerro	8/19/08	PAN0	Y	CU-843
961	LR-1271-06	FSL R8-2640	52	Bov non-clin	Cerro	8/19/08	PAN0	N	
1074	LR-1294-01	FSL R8-2824	52	Environmental	Cerro	10/8/08	PAN0	N	
1075	LR-1294-02	FSL R8-2825	52	Environmental	Cerro	10/8/08	PAN0	N	
1076	LR-1294-03	FSL R8-2826	52	Environmental	Cerro	10/8/08	PAN0	Y	CU-843
1169	LR-1332-01	FSL R8-3141	52	Environmental	Cerro	12/15/08	PAN0	Y	CU-843
1170	LR-1332-02	FSL R8-3142	52	Environmental	Cerro	12/15/08	PAN0	N	
1171	LR-1332-03	FSL R8-3143	52	Environmental	Cerro	12/15/08	PAN0	N	
1274	LR-1360-01	FSL R8-3432	52	Environmental	Cerro	2/14/09	PAN0	N	
1275	LR-1360-02	FSL R8-3433	52	Environmental	Cerro	2/14/09	PAN0	Y	CU-843
1276	LR-1360-03	FSL R8-3434	52	Environmental	Cerro	2/14/09	PAN0	N	
1101	LR-1309-01	FSL R8-2851	53	Environmental	Kentucky	11/6/08	PAN0	Y	CU-96
1115	LR-1317-01	FSL R8-2865	53	Environmental	Kentucky	12/8/08	PAN0	Y	CU-96
1211	LR-1342-01	FSL R8-3183	53	Bov non-clin	Kentucky	12/8/08	PAN0	N	
1212	LR-1342-02	FSL R8-3184	53	Bov non-clin	Kentucky	12/8/08	PAN0	N	
1213	LR-1342-03	FSL R8-3185	53	Bov non-clin	Kentucky	12/8/08	PAN0	Y	CU-96
1214	LR-1342-04	FSL R8-3186	53	Bov non-clin	Kentucky	12/8/08	PAN0	N	
1215	LR-1342-05	FSL R8-3187	53	Bov non-clin	Kentucky	12/8/08	PAN0	N	
1216	LR-1342-06	FSL R8-3188	53	Bov non-clin	Kentucky	12/8/08	PAN0	N	
1185	LR-1339-01	FSL R8-3157	53	Environmental	Kentucky	1/20/09	PAN0	N	
1186	LR-1339-02	FSL R8-3158	53	Environmental	Kentucky	1/20/09	PAN0	Y	CU-96
1187	LR-1340-01	FSL R8-3159	53	Environmental	Cerro* (Kentucky)	1/20/09	PAN0	Y	CU-213

Appendix Table 1

1293	LR-1367-01	FSL R8-3451	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1294	LR-1367-02	FSL R8-3452	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1295	LR-1367-03	FSL R8-3453	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1296	LR-1367-04	FSL R8-3454	53	Bov non-clin	Kentucky	1/20/09	PAN0	Y	CU-96
1297	LR-1367-05	FSL R8-3455	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1298	LR-1367-06	FSL R8-3456	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1299	LR-1367-07	FSL R8-3457	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1300	LR-1367-08	FSL R8-3458	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1301	LR-1367-09	FSL R8-3459	53	Bov non-clin	Kentucky	1/20/09	PAN0	N	
1281	LR-1362-01	FSL R8-3439	53	Environmental	Kentucky	3/3/09	PAN0	Y	CU-96
1282	LR-1362-02	FSL R8-3440	53	Environmental	Kentucky	3/3/09	PAN0	N	
1283	LR-1362-03	FSL R8-3441	53	Environmental	Kentucky	3/3/09	PAN0	N	
1327	LR-1369-01	FSL R8-3485	53	Bov non-clin	Kentucky	3/3/09	PAN0	Y	CU-96
1328	LR-1369-02	FSL R8-3486	53	Bov non-clin	Kentucky	3/3/09	PAN0	N	
1329	LR-1369-03	FSL R8-3487	53	Bov non-clin	Kentucky	3/3/09	PAN0	N	
1106	LR-1313-01	FSL R8-2856	54	Environmental	Cerro	11/26/08	PAN0	Y	CU-840
1061	LR-1287-01	FSL R8-2811	55	Environmental	Infantis	9/29/08	PAN0	Y	CU-107
1348	LR-1380-01	FSL R8-3666	55	Environmental	Cerro	1/14/09	PAN0	N	
1349	LR-1380-02	FSL R8-3667	55	Environmental	Cerro	1/14/09	PAN0	Y	CU-213
1288	LR-1365-01	FSL R8-3446	55	Environmental	Cerro	3/12/09	PAN0	Y	CU-213
1289	LR-1365-02	FSL R8-3447	55	Environmental	Cerro	3/12/09	PAN0	N	
110	LR-1057-01	FSL R8-349	56	Environmental	Anatum	2/22/08	PAN0	N	
111	LR-1057-02	FSL R8-350	56	Environmental	Anatum	2/22/08	PAN0	Y	CU-430
112	LR-1057-03	FSL R8-351	56	Environmental	Anatum	2/22/08	PAN0	N	
357	LR-1144-01	FSL R8-907	56	Environmental	Anatum	4/18/08	PAN0	Y	CU-430
358	LR-1144-02	FSL R8-908	56	Environmental	Anatum	4/18/08	PAN0	N	
418	LR-1160-01	FSL R8-968	56	Bov non-clin	Anatum	4/19/08	TEL1	Y	CU-430
419	LR-1161-01	FSL R8-969	56	Bov non-clin	Anatum	4/19/08	PAN0	Y	CU-430
420	LR-1161-02	FSL R8-970	56	Bov non-clin	Anatum	4/19/08	PAN0	N	
528	LR-1178-01	FSL R8-1401	56	Environmental	Anatum	6/12/08	AMC1 AMP1 FOX1 CEF1 CRO2	N	
529	LR-1178-02	FSL R8-1402	56	Environmental	Anatum	6/12/08	AMC1 AMP1 FOX1 CEF1 CRO2	Y	CU-1026
1334	LR-1372-01	FSL R8-3652	56	Environmental	Anatum* (Not serotyped)	3/25/09	CHL2 STR1	Y	CU-430
1335	LR-1373-01	FSL R8-3653	56	Environmental	Anatum* (Not serotyped)	3/25/09	CEF1 SUL1	Y	CU-430
338	LR-1134-01	FSL R8-888	57	Environmental	Cerro	3/19/08	PAN0	N	
339	LR-1134-02	FSL R8-889	57	Environmental	Cerro	3/19/08	PAN0	Y	CU-213
340	LR-1134-03	FSL R8-890	57	Environmental	Cerro	3/19/08	PAN0	N	
352	LR-1141-01	FSL R8-902	57	Environmental	Cerro	4/15/08	PAN0	N	
353	LR-1141-02	FSL R8-903	57	Environmental	Cerro	4/15/08	PAN0	Y	CU-213
354	LR-1142-01	FSL R8-904	57	Environmental	Cerro* (Untypeable)	4/15/08	PAN0	Y	CU-213
422	LR-1163-01	FSL R8-972	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
423	LR-1163-02	FSL R8-973	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
424	LR-1163-03	FSL R8-974	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
425	LR-1163-04	FSL R8-975	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
426	LR-1163-05	FSL R8-976	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
427	LR-1163-06	FSL R8-977	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
429	LR-1163-07	FSL R8-979	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
430	LR-1163-08	FSL R8-980	57	Bov non-clin	Cerro	4/28/08	PAN0	Y	CU-213
431	LR-1163-09	FSL R8-981	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
432	LR-1163-10	FSL R8-982	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
433	LR-1163-11	FSL R8-983	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
434	LR-1163-12	FSL R8-984	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
435	LR-1163-13	FSL R8-985	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
436	LR-1163-14	FSL R8-986	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
437	LR-1163-15	FSL R8-987	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
438	LR-1163-16	FSL R8-988	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
439	LR-1164-02	FSL R8-989	57	Bov non-clin	Cerro* (Untypeable Rough O:z4,z23:-)	4/28/08	PAN0	Y	CU-213
440	LR-1163-17	FSL R8-990	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
442	LR-1163-18	FSL R8-992	57	Bov non-clin	Cerro	4/28/08	PAN0	N	

Appendix Table 1

443	LR-1163-19	FSL R8-993	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
444	LR-1163-20	FSL R8-994	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
445	LR-1163-21	FSL R8-995	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
446	LR-1163-22	FSL R8-996	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
447	LR-1163-23	FSL R8-997	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
448	LR-1163-24	FSL R8-998	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
449	LR-1163-25	FSL R8-999	57	Bov non-clin	Cerro	4/28/08	PAN0	N	
535	LR-1184-01	FSL R8-1408	57	Environmental	Cerro	6/16/08	PAN0	N	
536	LR-1184-02	FSL R8-1409	57	Environmental	Cerro	6/16/08	PAN0	N	
537	LR-1184-03	FSL R8-1410	57	Environmental	Cerro	6/16/08	PAN0	N	
538	LR-1184-04	FSL R8-1411	57	Environmental	Cerro	6/16/08	PAN0	Y	CU-213
639	LR-1212-01	FSL R8-1648	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
640	LR-1212-02	FSL R8-1649	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
641	LR-1212-03	FSL R8-1650	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
642	LR-1212-04	FSL R8-1651	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
643	LR-1212-05	FSL R8-1652	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
644	LR-1212-06	FSL R8-1653	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
645	LR-1212-07	FSL R8-1654	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
646	LR-1212-08	FSL R8-1655	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
647	LR-1212-09	FSL R8-1656	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
648	LR-1212-10	FSL R8-1657	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
649	LR-1212-11	FSL R8-1658	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
650	LR-1212-12	FSL R8-1659	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
651	LR-1212-13	FSL R8-1660	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
652	LR-1212-14	FSL R8-1661	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
653	LR-1212-15	FSL R8-1662	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
654	LR-1212-16	FSL R8-1663	57	Bov non-clin	Cerro	6/16/08	PAN0	Y	CU-213
655	LR-1212-17	FSL R8-1664	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
656	LR-1212-18	FSL R8-1665	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
657	LR-1212-19	FSL R8-1666	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
658	LR-1212-20	FSL R8-1667	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
659	LR-1212-21	FSL R8-1668	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
660	LR-1212-22	FSL R8-1669	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
661	LR-1212-23	FSL R8-1670	57	Bov non-clin	Cerro	6/16/08	PAN0	N	
619	LR-1201-01	FSL R8-1627	57	Environmental	Cerro	7/21/08	PAN0	N	
620	LR-1201-02	FSL R8-1628	57	Environmental	Cerro (Typhimurium)	7/21/08	PAN0	Y	CU-213
621	LR-1201-03	FSL R8-1629	57	Environmental	Cerro	7/21/08	PAN0	N	
900	LR-1261-01	FSL R8-2364	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
901	LR-1261-02	FSL R8-2365	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
902	LR-1261-03	FSL R8-2366	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
903	LR-1261-04	FSL R8-2367	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
904	LR-1261-05	FSL R8-2368	57	Bov non-clin	Cerro	7/21/08	PAN0	Y	CU-213
905	LR-1261-06	FSL R8-2369	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
906	LR-1261-07	FSL R8-2370	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
907	LR-1261-08	FSL R8-2371	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
908	LR-1261-09	FSL R8-2372	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
909	LR-1261-10	FSL R8-2373	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
910	LR-1261-11	FSL R8-2374	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
911	LR-1261-12	FSL R8-2375	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
912	LR-1261-13	FSL R8-2376	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
913	LR-1261-14	FSL R8-2377	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
914	LR-1261-15	FSL R8-2378	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
915	LR-1261-16	FSL R8-2379	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
916	LR-1261-17	FSL R8-2380	57	Bov non-clin	Cerro	7/21/08	PAN0	N	
768	LR-1228-01	FSL R8-2011	57	Environmental	Cerro	8/26/08	PAN0	Y	CU-213
769	LR-1228-02	FSL R8-2012	57	Environmental	Cerro	8/26/08	PAN0	N	

Appendix Table 1

869	LR-1257-01	FSL R8-2245	57	Environmental	Thompson* (Cerro)	9/30/08	PAN0	Y	CU-157
870	LR-1258-01	FSL R8-2246	57	Environmental	Cerro	9/30/08	PAN0	Y	CU-213
871	LR-1258-02	FSL R8-2247	57	Environmental	Cerro	9/30/08	PAN0	N	
1087	LR-1302-01	FSL R8-2837	57	Environmental	Cerro	11/4/08	PAN0	Y	CU-213
1088	LR-1302-02	FSL R8-2838	57	Environmental	Cerro	11/4/08	PAN0	N	
1089	LR-1302-03	FSL R8-2839	57	Environmental	Cerro	11/4/08	PAN0	N	
1112	LR-1316-01	FSL R8-2862	57	Environmental	Cerro	12/9/08	PAN0	Y	CU-213
1113	LR-1316-02	FSL R8-2863	57	Environmental	Cerro	12/9/08	PAN0	N	
1114	LR-1316-03	FSL R8-2864	57	Environmental	Cerro	12/9/08	PAN0	N	
1175	LR-1335-01	FSL R8-3147	57	Environmental	Cerro	1/6/09	PAN0	N	
1176	LR-1335-02	FSL R8-3148	57	Environmental	Cerro	1/6/09	PAN0	Y	CU-213
1177	LR-1335-03	FSL R8-3149	57	Environmental	Cerro	1/6/09	PAN0	N	
1267	LR-1357-01	FSL R8-3425	57	Environmental	Cerro	2/17/09	PAN0	N	
1268	LR-1357-02	FSL R8-3426	57	Environmental	Cerro	2/17/09	PAN0	N	
1269	LR-1357-03	FSL R8-3427	57	Environmental	Cerro	2/17/09	PAN0	Y	CU-213
1290	LR-1366-01	FSL R8-3448	57	Environmental	Cerro	3/24/09	PAN0	N	
1291	LR-1366-02	FSL R8-3449	57	Environmental	Cerro	3/24/09	PAN0	N	
1292	LR-1366-03	FSL R8-3450	57	Environmental	Cerro	3/24/09	PAN0	Y	CU-213
786	LR-1240-01	FSL R8-2030	59	Environmental	Cerro	Not given	PAN0	Y	CU-213
541	LR-1187-01	FSL R8-1414	60	Environmental	Cerro	6/17/08	PAN0	N	
542	LR-1187-02	FSL R8-1415	60	Environmental	Cerro	6/17/08	PAN0	Y	CU-973
662	LR-1213-01	FSL R8-1671	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
663	LR-1213-02	FSL R8-1672	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
664	LR-1213-03	FSL R8-1673	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
665	LR-1213-04	FSL R8-1674	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
666	LR-1213-05	FSL R8-1675	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
667	LR-1213-06	FSL R8-1676	60	Bov non-clin	Cerro (Newport)	6/17/08	PAN0	E	
668	LR-1213-07	FSL R8-1677	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
669	LR-1213-08	FSL R8-1678	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
670	LR-1213-09	FSL R8-1679	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
671	LR-1213-10	FSL R8-1680	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
672	LR-1213-11	FSL R8-1681	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
673	LR-1213-12	FSL R8-1682	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
674	LR-1214-01	FSL R8-1683	60	Bov non-clin	Newport	6/17/08	PAN0	Y	CU-1017
675	LR-1213-13	FSL R8-1684	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
676	LR-1213-14	FSL R8-1685	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
798	LR-1213-15	FSL R8-2042	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
799	LR-1213-16	FSL R8-2043	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
800	LR-1213-17	FSL R8-2044	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
801	LR-1213-18	FSL R8-2045	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
802	LR-1213-19	FSL R8-2046	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
803	LR-1213-20	FSL R8-2047	60	Bov non-clin	Cerro* (Thompson)	6/17/08	PAN0	E	
804	LR-1213-21	FSL R8-2048	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
805	LR-1213-22	FSL R8-2049	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
806	LR-1213-23	FSL R8-2050	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
807	LR-1213-24	FSL R8-2051	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
808	LR-1213-25	FSL R8-2052	60	Bov non-clin	Cerro	6/17/08	PAN0	Y	CU-213
809	LR-1213-26	FSL R8-2053	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
810	LR-1213-27	FSL R8-2054	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
811	LR-1213-28	FSL R8-2055	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
812	LR-1213-29	FSL R8-2056	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
813	LR-1213-30	FSL R8-2057	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
814	LR-1213-31	FSL R8-2058	60	Bov non-clin	Cerro	6/17/08	PAN0	N	
917	LR-1262-01	FSL R8-2381	60	Bov non-clin	Typhimurium Copenhagen	7/22/08	AMC1 AMP1 CHL1 STR1 SUL1 TEL1	Y	CU-1171
918	LR-1263-01	FSL R8-2382	60	Bov non-clin	Cerro	7/22/08	PAN0	N	
919	LR-1263-02	FSL R8-2383	60	Bov non-clin	Cerro	7/22/08	PAN0	N	
920	LR-1263-03	FSL R8-2384	60	Bov non-clin	Cerro	7/22/08	PAN0	N	

Appendix Table 1

921	LR-1263-04	FSL R8-2385	60	Bov non-clin	Cerro	7/22/08	PAN0	Y	CU-213
778	LR-1235-01	FSL R8-2022	60	Environmental	Cerro	9/4/08	PAN0	Y	CU-213
970	LR-1275-01	FSL R8-2649	60	Bov non-clin	Newport	9/4/08	PAN0	Y	CU-1014
1099	LR-1307-01	FSL R8-2849	60	Environmental	Montevideo	11/10/08	PAN0	Y	CU-1006
1100	LR-1308-01	FSL R8-2850	60	Environmental	Cerro	11/10/08	PAN0	Y	CU-213
1259	LR-1353-01	FSL R8-3417	60	Environmental	Montevideo	1/20/09	PAN0	Y	CU-1006
1260	LR-1354-01	FSL R8-3418	60	Environmental	Cerro	1/20/09	PAN0	Y	CU-213
1340	LR-1376-01	FSL R8-3658	60	Environmental	Montevideo	4/13/09	PAN0	N	
1341	LR-1376-02	FSL R8-3659	60	Environmental	Montevideo	4/13/09	PAN0	Y	CU-1006
1342	LR-1377-01	FSL R8-3660	60	Environmental	Cerro	4/13/09	PAN0	Y	CU-213
539	LR-1185-01	FSL R8-1412	61	Environmental	Cerro	6/17/08	AMC1 AMP1 CHL2 TEL1	Y	CU-213
540	LR-1186-01	FSL R8-1413	61	Environmental	Cerro	6/17/08	PAN0	Y	CU-213
922	LR-1264-01	FSL R8-2386	61	Bov non-clin	Cerro	7/23/08	PAN0	N	
923	LR-1264-02	FSL R8-2387	61	Bov non-clin	Cerro	7/23/08	PAN0	N	
924	LR-1264-03	FSL R8-2388	61	Bov non-clin	Cerro	7/23/08	PAN0	Y	CU-213
779	LR-1236-01	FSL R8-2023	61	Environmental	Cerro	9/3/08	PAN0	N	
780	LR-1236-02	FSL R8-2024	61	Environmental	Cerro	9/3/08	PAN0	Y	CU-213
962	LR-1272-01	FSL R8-2641	61	Bov non-clin	Cerro	9/3/08	PAN0	N	
963	LR-1272-02	FSL R8-2642	61	Bov non-clin	Cerro	9/3/08	PAN0	Y	CU-213
964	LR-1272-03	FSL R8-2643	61	Bov non-clin	Cerro	9/3/08	PAN0	N	
965	LR-1272-04	FSL R8-2644	61	Bov non-clin	Cerro	9/3/08	PAN0	N	
966	LR-1272-05	FSL R8-2645	61	Bov non-clin	Cerro	9/3/08	PAN0	N	
1119	LR-1320-01	FSL R8-2869	61	Bov non-clin	Cerro	10/14/08	PAN0	Y	CU-213
1120	LR-1320-02	FSL R8-2870	61	Bov non-clin	Cerro	10/14/08	PAN0	N	
1121	LR-1320-03	FSL R8-2871	61	Bov non-clin	Cerro	10/14/08	PAN0	N	
1261	LR-1355-01	FSL R8-3419	61	Environmental	Cerro	1/29/09	PAN0	Y	CU-213
1262	LR-1355-02	FSL R8-3420	61	Environmental	Cerro	1/29/09	PAN0	N	
1263	LR-1355-03	FSL R8-3421	61	Environmental	Cerro	1/29/09	PAN0	N	
1343	LR-1378-01	FSL R8-3661	61	Environmental	Cerro	4/12/09	PAN0	Y	CU-213
1344	LR-1378-02	FSL R8-3662	61	Environmental	Cerro	4/12/09	PAN0	N	
623	LR-1203-01	FSL R8-1631	62	Environmental	Cerro	7/22/08	CHL2 TEL1	Y	CU-213
624	LR-1204-01	FSL R8-1632	62	Environmental	Cerro	7/22/08	PAN0	Y	CU-213
781	LR-1237-01	FSL R8-2025	62	Environmental	Minnesota	9/3/08	PAN0	Y	CU-523
782	LR-1238-01	FSL R8-2026	62	Environmental	Cerro	9/3/08	PAN0	Y	CU-213
783	LR-1238-02	FSL R8-2027	62	Environmental	Cerro (Minnesota)	9/3/08	PAN0	E	
967	LR-1273-01	FSL R8-2646	62	Bov non-clin	Cerro	9/3/08	PAN0	Y	CU-213
968	LR-1273-02	FSL R8-2647	62	Bov non-clin	Cerro	9/3/08	PAN0	N	
969	LR-1274-01	FSL R8-2648	62	Bov non-clin	Minnesota* (Rubislaw)	9/3/08	PAN0	Y	CU-523
1067	LR-1290-01	FSL R8-2817	62	Environmental	Cerro	10/14/08	PAN0	N	
1068	LR-1290-02	FSL R8-2818	62	Environmental	Cerro	10/14/08	PAN0	Y	CU-213
1122	LR-1321-01	FSL R8-2872	62	Bov non-clin	Cerro	10/14/08	PAN0	Y	CU-213
1123	LR-1322-01	FSL R8-2873	62	Bov non-clin	Newport	10/14/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
1124	LR-1322-02	FSL R8-2874	62	Bov non-clin	Newport	10/14/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
1102	LR-1310-01	FSL R8-2852	62	Environmental	Cerro	11/12/08	PAN0	Y	CU-213
1149	LR-1325-01	FSL R8-2899	62	Bov non-clin	Newport	11/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
1150	LR-1325-02	FSL R8-2900	62	Bov non-clin	Newport	11/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
1151	LR-1325-03	FSL R8-2901	62	Bov non-clin	Newport	11/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
1152	LR-1326-01	FSL R8-2902	62	Bov non-clin	Cerro	11/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
1153	LR-1326-02	FSL R8-2903	62	Bov non-clin	Cerro	11/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-213
1154	LR-1326-03	FSL R8-2904	62	Bov non-clin	Cerro	11/12/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	N	
1165	LR-1329-01	FSL R8-3137	62	Environmental	Newport	12/11/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 STR1 SUL1 TEL1	Y	CU-121
1166	LR-1330-01	FSL R8-3138	62	Environmental	Cerro	12/11/08	PAN0	N	
1167	LR-1331-01	FSL R8-3139	62	Environmental	Newport	12/11/08	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
1168	LR-1330-02	FSL R8-3140	62	Environmental	Cerro	12/11/08	PAN0	Y	CU-213
1252	LR-1349-01	FSL R8-3410	62	Environmental	Newport	1/22/09	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
1253	LR-1350-01	FSL R8-3411	62	Environmental	Cerro (Multiple serovars)	1/22/09	PAN0	E	
1254	LR-1350-02	FSL R8-3412	62	Environmental	Cerro	1/22/09	PAN0	N	
1255	LR-1350-03	FSL R8-3413	62	Environmental	Cerro	1/22/09	PAN0	Y	CU-213
1284	LR-1363-01	FSL R8-3442	62	Environmental	Newport (Minnesota)	3/5/09	PAN0	Y	CU-121
1285	LR-1364-01	FSL R8-3443	62	Environmental	Cerro	3/5/09	PAN0	N	

Appendix Table 1

1286	LR-1364-02	FSL R8-3444	62	Environmental	Cerro	3/5/09	PAN0	N	
1287	LR-1364-03	FSL R8-3445	62	Environmental	Cerro	3/5/09	PAN0	Y	CU-213
1345	LR-1379-01	FSL R8-3663	62	Environmental	Cerro	4/16/09	PAN0	N	
1346	LR-1379-02	FSL R8-3664	62	Environmental	Cerro	4/16/09	PAN0	Y	CU-213
1347	LR-1379-03	FSL R8-3665	62	Environmental	Cerro	4/16/09	PAN0	N	
1352	LR-1382-01	FSL R8-4025	62	Environmental	Cerro	5/28/09	PAN0	Y	CU-213
1353	LR-1383-01	FSL R8-4026	62	Environmental	Newport	5/28/09	AMC1 AMP1 FOX1 CEF1 CRO2 CHL1 KAN1 STR1 SUL1 TEL1	Y	CU-121
1354	LR-1382-02	FSL R8-4027	62	Environmental	Cerro	5/28/09	PAN0	N	
1356	LR-1385-01	FSL R8-4029	62	Environmental	Muenster	7/9/09	PAN0	Y	CU-007
1357	LR-1386-01	FSL R8-4030	62	Environmental	Cerro	7/9/09	PAN0	Y	CU-213
866	LR-1255-01	FSL R8-2242	64	Environmental	Orion var. 15+, 34+	9/27/08	PAN0	Y	CU-175
1270	LR-1358-01	FSL R8-3428	64	Environmental	Orion var. 15+, 34+	2/17/09	PAN0	Y	CU-1028
792	LR-1245-01	FSL R8-2036	65	Environmental	Cerro	9/12/08	PAN0	Y	CU-213
793	LR-1245-02	FSL R8-2037	65	Environmental	Cerro	9/12/08	PAN0	N	
794	LR-1245-03	FSL R8-2038	65	Environmental	Cerro	9/12/08	PAN0	N	
971	LR-1276-01	FSL R8-2650	65	Bov non-clin	Kentucky* (Untypeable Rough O:i:z6)	9/12/08	PAN0	Y	CU-96
972	LR-1277-01	FSL R8-2651	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
973	LR-1277-02	FSL R8-2652	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
974	LR-1277-03	FSL R8-2653	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
975	LR-1277-04	FSL R8-2654	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
976	LR-1277-05	FSL R8-2655	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
977	LR-1277-06	FSL R8-2656	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
978	LR-1277-07	FSL R8-2657	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
979	LR-1277-08	FSL R8-2658	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
980	LR-1277-09	FSL R8-2659	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
981	LR-1277-10	FSL R8-2660	65	Bov non-clin	Cerro	9/12/08	PAN0	Y	CU-213
982	LR-1277-11	FSL R8-2661	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
983	LR-1277-12	FSL R8-2662	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
984	LR-1277-13	FSL R8-2663	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
985	LR-1277-14	FSL R8-2664	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
986	LR-1277-15	FSL R8-2665	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
987	LR-1277-16	FSL R8-2666	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
988	LR-1277-17	FSL R8-2667	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
989	LR-1277-18	FSL R8-2668	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
990	LR-1277-19	FSL R8-2669	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
991	LR-1277-20	FSL R8-2670	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
992	LR-1277-21	FSL R8-2671	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
993	LR-1277-22	FSL R8-2672	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
994	LR-1277-23	FSL R8-2673	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
995	LR-1277-24	FSL R8-2674	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
996	LR-1277-25	FSL R8-2675	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
997	LR-1277-26	FSL R8-2676	65	Bov non-clin	Cerro* (Not serotyped)	9/12/08	PAN0	E	
998	LR-1277-27	FSL R8-2677	65	Bov non-clin	Cerro	9/12/08	PAN0	N	
1072	LR-1292-01	FSL R8-2822	65	Environmental	Cerro	10/15/08	PAN0	Y	CU-213
1073	LR-1293-01	FSL R8-2823	65	Environmental	Cerro	10/15/08	AMC1 AMP1 CEF2 SUL1 TEL2	Y	CU-213
1125	LR-1323-01	FSL R8-2875	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1126	LR-1323-02	FSL R8-2876	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1127	LR-1323-03	FSL R8-2877	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1128	LR-1323-04	FSL R8-2878	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1129	LR-1323-05	FSL R8-2879	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1130	LR-1323-06	FSL R8-2880	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1131	LR-1323-07	FSL R8-2881	65	Bov non-clin	Cerro	10/15/08	PAN0	Y	CU-213
1132	LR-1323-08	FSL R8-2882	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1133	LR-1323-09	FSL R8-2883	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1134	LR-1323-10	FSL R8-2884	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1135	LR-1323-11	FSL R8-2885	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1136	LR-1323-12	FSL R8-2886	65	Bov non-clin	Cerro	10/15/08	PAN0	N	

Appendix Table 1

1137	LR-1323-13	FSL R8-2887	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1138	LR-1323-14	FSL R8-2888	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1139	LR-1323-15	FSL R8-2889	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1140	LR-1323-16	FSL R8-2890	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1141	LR-1323-17	FSL R8-2891	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1142	LR-1323-18	FSL R8-2892	65	Bov non-clin	Cerro	10/15/08	PAN0	N	
1103	LR-1311-01	FSL R8-2853	65	Environmental	Cerro	11/12/08	PAN0	N	
1104	LR-1311-02	FSL R8-2854	65	Environmental	Cerro	11/12/08	PAN0	Y	CU-213
1105	LR-1312-01	FSL R8-2855	65	Environmental	Cerro* (Not serotyped)	11/12/08	AMP2	Y	CU-213
1155	LR-1327-01	FSL R8-2905	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1156	LR-1327-02	FSL R8-2906	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1157	LR-1327-03	FSL R8-2907	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1158	LR-1327-04	FSL R8-2908	65	Bov non-clin	Cerro	11/13/08	PAN0	Y	CU-213
1159	LR-1327-05	FSL R8-2909	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1160	LR-1327-06	FSL R8-2910	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1161	LR-1327-07	FSL R8-2911	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1162	LR-1327-08	FSL R8-2912	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1163	LR-1327-09	FSL R8-2913	65	Bov non-clin	Cerro	11/13/08	PAN0	N	
1180	LR-1337-01	FSL R8-3152	65	Environmental	Cerro	1/6/09	PAN0	N	
1181	LR-1337-02	FSL R8-3153	65	Environmental	Cerro	1/6/09	PAN0	Y	CU-213
1271	LR-1359-01	FSL R8-3429	65	Environmental	Cerro	2/17/09	PAN0	N	
1272	LR-1359-02	FSL R8-3430	65	Environmental	Cerro	2/17/09	PAN0	Y	CU-213
1273	LR-1359-03	FSL R8-3431	65	Environmental	Cerro	2/17/09	PAN0	N	
1337	LR-1375-01	FSL R8-3655	65	Environmental	Cerro	3/31/09	PAN0	N	
1338	LR-1375-02	FSL R8-3656	65	Environmental	Cerro	3/31/09	PAN0	N	
1339	LR-1375-03	FSL R8-3657	65	Environmental	Cerro	3/31/09	PAN0	Y	CU-213
1350	LR-1381-01	FSL R8-4023	65	Environmental	Cerro	5/26/09	PAN0	Y	CU-213
1351	LR-1381-02	FSL R8-4024	65	Environmental	Cerro	5/26/09	PAN0	N	
1355	LR-1384-01	FSL R8-4028	65	Environmental	Cerro	7/7/09	PAN0	Y	CU-213
1359	LR-1388-01	FSL R8-4032	65	Environmental	Cerro	8/11/09	PAN0	Y	CU-974

[a]Shaded cells indicate isolates for which PFGE-predicted serovar did not match the serovar reported based on classical serotyping (shown in parenthesis); in all cases, isolates with the same PFGE type isolated from the same farm showed, based on classical serotyping, the predicted PFGE serovar for that isolates. Hence, the classical serovar for these isolates were considered a misclassification and the serovars for these isolates were changed to the serovar predicted, by PFGE, for the given isolate. Serovar designations with an * are based on molecular serotyping data (see Supp. Table 2 for details).

[b] Y= yes; N=no; isolates classified as "E", in this column, were eliminated from the final PFGE type analysis because once their serovars were reclassified after additional analyses they were no longer representative isolates since there were other isolates obtained on the same date from the same farm and source that had the same antimicrobial susceptibility types.

Appendix Table 2. *Salmonella* isolates for which additional serovar analyses were performed[a]

FSL #	Farm No.	Serovar initially assigned based on traditional serotyping	Serovar prodicted based on PFGE type[b]	Molecular serotyping-predicted serovar	Final serovar
				Confirmed serovar by PCR	
Isolates for which serovar was changed based on PFGE-based serovar prediction and presence of the same serovar/PFGE type on the source farm for the isolate[c]					
FSL R8-346	1	Kentucky	Meleagridis	N/A	Meleagridis
FSL R8-2233	1	Multiple serovars	Meleagridis	N/A	Meleagridis
FSL R8-1417	10	Cerro	Oranienburg	N/A	Oranienburg
FSL R6-1000	17	Newport	Kentucky	N/A	Kentucky
FSL R8-342	17	Meleagridis	Newport	N/A	Newport
FSL R8-830	17	Kentucky	Newport	N/A	Newport
FSL R8-1418	18	Oranienburg	Cerro	N/A	Cerro
FSL R6-779	26	Cerro	Kentucky	N/A	Kentucky
FSL R8-1396	26	Kentucky	Cerro	N/A	Cerro
FSL R6-782	30	Multiple serovars	Cerro	N/A	Cerro
FSL R8-2007	52	Cerro	Mbandaka	N/A	Mbandaka
FSL R8-1628	57	Typhimurium	Cerro	N/A	Cerro
FSL R8-1676	60	Newport	Cerro	N/A	Cerro
FSL R8-2027	62	Minnesota	Cerro	N/A	Cerro
FSL R8-3411	62	Multiple serovars	Cerro	N/A	Cerro
FSL R8-3442	62	Minnesota	Newport	N/A	Newport
Isolates for which serovar is based on convergance of both PFGE-based serovar prediction and molecular serotyping					
FSL R6-799	14	Kentucky	Newport	Newport	Newport
FSL R8-208	17	Cerro	Typhimurium	Typhimurium	Typhimurium
FSL R8-3194	47	Lexington	Muenster	3,10:-:1,5	3,10:-:1,5
FSL R8-915	52	Kentucky	4,5,12:i:- *	4,5,12:i:-	4,5,12:i:-
FSL R8-3159	53	Kentucky	Cerro	Cerro	Cerro
FSL R8-2245	57	Cerro	Thompson	Thompson	Thompson
FSL R8-2047	60	Thompson	Cerro	Cerro	Cerro
FSL R8-2648	62	Rubislaw	Minnesota	Minnesota	Minnesota
Isolates that were classifed as untypeable by traditional as serotyping					
FSL R8-2829	1	Untypeable	Meleagridis	Meleagridis	Meleagridis
FSL R8-835	15	Untypeable; Rough O: Nonmotile	Dublin	Dublin	Dublin
FSL R8-1485	16	Untypeable Rough O:i:z6	Kentucky	Kentucky	Kentucky
FSL R8-2031	16	Untypeable; Rough O: Nonmotile	Kentucky*	Kentucky	Kentucky
FSL R6-962	19	Untypeable	Anatum	Anatum	Anatum
FSL R8-2860	39	Untypeable	Anatum	Anatum	Anatum
FSL R8-2236	42	Untypeable	Kentucky	Kentucky	Kentucky
FSL R8-904	57	Untypeable	Cerro	Cerro	Cerro
FSL R8-989	57	Untypeable; Rough O:z4,z23:-	Cerro	Cerro	Cerro
FSL R8-2650	65	Untypeable Rough O:i:z6	Kentucky	Kentucky	Kentucky
Isolates that were only characterized by PFGE and molecular serotyping					
FSL R8-2035	47	Not Serotyped	Muenster	3,10:-:1,5	3,10:-:1,5
FSL R8-3652	56	Not Serotyped	Anatum	Anatum	Anatum
FSL R8-3653	56	Not Serotyped	Anatum	Anatum	Anatum
FSL R8-2855	65	Not Serotyped	Cerro	Cerro	Cerro
FSL R8-2676	65	Not Serotyped	Cerro	Cerro	Cerro

[a]For 8 isolates traditional serovar and molecular type data matched even though isolates with closely related PFGE types represented a different serovar; these isolates are not included here

[b]PFGE data for all isolates were used to identify isolates in the FSL database that matched the given PFGE type; if multiple isolates with a matching PFGE type had been assigned a different serovar, this serovar is listed as "Serovar predicted based on PFGE type"

[c]For isolates where additional isolates with the same PFGE type, isolated from the same farm as a given isolate, showed, based on classical serotyping, the predicted PFGE serovar for that isolate, the given isolate was classified as the serovar predicted by PFGE (without additional confirmation)

Appendix Table 3. Antimicrobial resistance types among the 90 *Salmonella* isolates with resistance to at least one antimicrobial.

Resistance Type[a]	Resistance[b]	Number of isolates with a given resistance type representing serovar									Total
		Typhimurium	Newport	Cerro	Kentucky	Anatum	Meleagridis	Agona	Untypeable	Other[c]	
	11 Resistances										
1	AMC1-AMP1-FOX1-CEF1-CRO2-CHL1-KAN1-STR1-SUL1-TEL1-SXT1							1			1
	10 Resistances										
2	AMC1-AMP1-FOX1-CEF1-CHL1-KAN1-STR1-SUL1-TEL1-SXT1							1			1
3	AMC1-AMP1-FOX1-CEF1-CRO1-CHL1-KAN1-STR1-SUL1-TEL1		1		1						2
4	AMC1-AMP1-FOX1-CEF1-CRO2-CHL1-KAN1-STR1-SUL1-TEL1	1	8	1							10
	9 Resistances										
5	AMC1-AMP1-FOX1-CEF1-CHL1-KAN1-STR1-SUL1-TEL1		2							1	3
6	AMC1-AMP1-FOX1-CEF1-CRO1-CHL1-NAL1-SUL1-TEL1	1									1
7	AMC1-AMP1-FOX1-CEF1-CRO2-CHL1-STR1-SUL1-TEL1		8								8
	8 Resistances										
8	AMC1-AMP1-FOX1-CEF1-CHL1-STR1-SUL1-TEL1		1								1
9	AMC1-AMP1-FOX1-CEF1-CRO2-KAN1-STR1-TEL1									1	1
10	AMC1-AMP1-FOX1-CEF1-CRO2-STR1-SUL1-TEL1		1								1
11	AMC2-AMP1-FOX1-CEF2-KAN1-STR1-SUL1-TEL1	1									1
	7 Resistances										
12	AMP1-CHL1-KAN1-STR1-SUL1-TEL1-SXT1								1		1
	6 Resistances										
13	AMC1-AMP1-CHL1-STR1-SUL1-TEL1	1									1
14	AMC1-AMP1-FOX1-CEF1-CRO2-TEL1					1					1
15	AMC1-AMP1-FOX2-CEF1-CHL2-SUL1	1									1
16	AMC1-AMP1-FOX2-CEF1-SUL1-TEL1	1									1
17	AMC1-AMP1-KAN1-STR1-SUL1-TEL1	1									1
18	AMC2-AMP1-FOX1-CEF1-STR1-TEL1									1	1
19	AMC2-AMP1-KAN1-STR1-SUL1-TEL1	1									1
	5 Resistances										
20	AMC1-AMP1-CEF1-CHL2-SUL1	1									1
21	AMC1-AMP1-CEF2-SUL1-TEL2			1							1
22	AMC1-AMP1-CHL2-GEN2-TEL1									1	1
23	AMC1-AMP1-FOX1-CEF1-CRO2	1				2				1	4
24	AMP1-KAN1-STR1-SUL1-TEL1	5			1						6
	4 Resistances										
25	AMC1-AMP1-CHL2-SUL1	1									1
26	AMC1-AMP1-CHL2-TEL1			1	2						3
27	AMC1-AMP1-FOX1-CEF1	1									1
28	AMC1-FOX1-CEF2-CHL1	1									1
29	AMC2-AMP1-CEF1-SUL1			1							1
	3 Resistances										
30	AMC1-AMP1-FOX1				1						1
31	AMC1-AMP1-TEL1			2					1		3
32	AMC1-AMP2-TEL1			1							1
33	AMP1-CEF1-CRO1	1					1				2
34	FOX1-CEF2-CHL1	1									1
	2 Resistances										
35	AMC2-SUL1			1							1
36	CEF1-CRO2	2									2
37	CEF1-SUL1					1					1
38	CHL2-STR1					1					1
39	CHL2-TEL1	1		1							2
40	FOX1-CEF1	1									1
41	FOX1-CHL1	1									1

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42	FOX1-CHL2		1								1
43	KAN1-TEL2									1	1
	1 Resistance										
44	AMP1			1		1					2
45	AMP2			1							1
46	CEF1						2				2
47	CEF2						1				1
48	CHL2				1					1	2
49	SUL1			1							1
50	TEL1			1	2	1					4

[a]A resistance type number was assigned to each distinct resistance type (representing a unique combination of resistances against the antimicrobials evaluated).

[b]Numbers following the antimicrobial abbreviations (e.g., AMP) indicate either resistance (number 1) or intermediate resistance (number 2) to the antimicrobial. Antimicrobial abbreviation legend: AMK=amikacin; AMC=amoxicillin/clavulanic acid; AMP=ampicillin; FOX=cefoxitin; CEF=ceftiofur; CRO=ceftriaxone; CHL=cloramphenicol; CIP=ciprofloxacin; GEN=gentamicin; KAN=kanamycin; NAL=nalidixic acid; STR=streptomycin; SUL=sulfisoxazole; TEL=tetracycline; SXT=trimethoprim/sulfamethoxazole.

[c]"Other" includes all serovars for which only 1 resistant isolates was identified, these include serovars 4,5,12:i– (resistance type 23); 3,10:–:1,5 (resistance type 43); Heidelberg (resistance type 9); Mbandaka (resistance type 18); Minnesota (resistance type 48); Muenster (resistance type 5); Oranienburg (resistance type 22).

Appendix Table 4. List of isolates used in invasion assays.

Serovar	Isolate ID	Source	Year of isolation
Cerro	R8-2280	Bovine clinical	2008
	R8-2660	Bovine non-clinical	2008
	R8-2827	Farm environment	2008
	R8-3972	Human clinical	2007
Kentucky	R8-3283	Bovine clinical	2008
	R8-083	Bovine non-clinical	2007
	R8-817	Farm environment	2008
	A4-831	Human*	2006
Newport	R8-4115	Bovine clinical	2009
	R8-829	Bovine non-clinical	2008
	R8-4026	Farm environment	2009
	R8-2926	Human clinical	2008
Typhimurium	R8-2185	Bovine clinical	2008
	R8-272	Bovine non-clinical	2008
	R8-171	Farm environment	2007
	R8-2481	Human clinical	2008

*The information provided by the New York State Department of Health (NYSDOH) for this isolate, did not specify if it was a clinical case

Appendix Table 5. Genomes and sequences used for cdtB, pltA, and pltB analyses[a]

		Genbank accessions				
Strain	**Clade in Fig. 1**	***cdtB***	**pltA (*cdtB* associated)**	***artA***	**pltB (*cdtB* associated)**	***artB***
Salmonella enterica subsp. enterica Urbana FSL R8-2977		AFCW01001088.1	ZP_12186254	AFCW01000865.1	ZP_12186255	AFCW01000865.1
Salmonella enterica subsp. enterica Typhi CT18		NP_456275	NP_456278	AL513382.1	NP_456279	NP_455807
Salmonella enterica subsp. enterica Typhi str. Ty2		AAO68774.1	AE014613.1	AE014613.1	AE014613.1	AE014613.1
Salmonella enterica subsp. enterica Javiana str. CFSAN001992		AGF81397.1	CP004027.1	YP_007470370	YP_007470839	CP004027.1
Salmonella enterica subsp. enterica Schwarzengrund str. CVM19633		ACF89261.1	YP_002114250	CP001127.1	CP001127.1	CP001127.1
Salmonella enterica subsp. enterica Schwarzengrund str. SL480		ABEJ01000026.1	ZP_02664208	ABEJ01000003.1	ABEJ01000026.1	ABEJ01000003.1
Salmonella enterica subsp. enterica Javiana str. GA_MM04042433		ABEH02000010.1	ZP_03221644	ABEH02000002.1	ABEH02000010.1	ABEH02000002.1
Salmonella enterica subsp. enterica Pomona str. ATCC 10729		AHIA01000037.1	AHIA01000037.1	AHIA01000012.1	AHIA01000037.1	ZP_13077485
Salmonella enterica subsp. enterica Montevideo str. 315996572		AESH01000006.1	ZP_11690104	AESH01000022.1	ZP_11690103	AESH01000022.1
Salmonella enterica subsp. enterica Give FSL S5-487		ZP_12133468	AFCM01000737.1	ZP_12132888	ZP_12133472	ZP_12132889
Salmonella enterica subsp. enterica Johannesburg FSL S5-703		ZP_12148338	AFCP01000578.1	AFCP01000828.1	AFCP01000578.1	AFCP01000828.1
Salmonella enterica subsp. enterica Montevideo FSL S5-403		ZP_12164725	AFCS01000632.1	ZP_12164178	AFCS01000632.1	AFCS01000509.1
Salmonella enterica subsp. enterica Gaminara FSL A4-567		ZP_12128237	AFCL01000868.1	ZP_12127714	AFCL01000868.1	ZP_12127715
Salmonella enterica subsp. enterica Rubislaw FSL A4-653		ZP_12170420	ZP_12170423	ZP_12169861.	ZP_12170424	AFCT01000901.1
Salmonella enterica subsp. enterica Minnesota FSL A4-603		ZP_12153324	AFCQ01000579	AFCQ01000963.1	AFCQ01000579.1	AFCQ01000963.1
Salmonella enterica subsp. enterica Paratyphi_A_str_ATCC_9150		YP_150845	YP_150848	CP000026.1	YP_150849	YP_150458
Salmonella enterica subsp. enterica Paratyphi A str. AKU_12601		CAR59673.1	YP_002142332	FM200053.1	FM200053.1	YP_002141943
Salmonella enterica subsp. arizonae_str_RSK2980		YP_001571391				
Salmonella enterica subsp. enterica Inverness_FSL R8-3668		ZP_12142538				
Salmonella bongori NCTC_12419		YP_004729962				
Haemophilus parasuis SH0165		YP_002474906				
Aggregatibacter_actinomycetemcomitans_RhAA1		ZP_13036112				
Aggregatibacter_actinomycetemcomitans_ANH9381		YP_004948954				
Aggregatibacter actinomycetemcomitans D11S-1		YP_003255523				
Aggregatibacter actinomycetemcomitans D7S-1		YP_006287827				
Aggregatibacter actinomycetemcomitans serotype b str. SCC1398		ZP_11585302				
Aggregatibacter actinomycetemcomitans serotype b str. SCC4092		ZP_21151980				
Aggregatibacter_actinomycetemcomitans_Y4		AAC70898				
Haemophilus ducreyi 35000HP		NP_873398				
Campylobacter fetus subsp. venerealis NCTC 10354	clade 2	ZP_18245823				
Campylobacter fetus subsp. venerealis str. Azul-94	clade 2	ZP_06009420				
Campylobacter fetus subsp. fetus 82-40	clade 2	YP_892124				
Campylobacter jejuni RM1221		YP_178098				
Campylobacter jejuni subsp. jejuni CF93-6		ZP_01067880				
Campylobacter jejuni subsp. jejuni CG8421		ZP_03223221				
Campylobacter jejuni subsp. jejuni NW		ZP_18271920				
Campylobacter jejuni subsp. jejuni 1997-11		ZP_14199461				
Campylobacter jejuni subsp. jejuni 81116		YP_001481648				
Campylobacter jejuni subsp. jejuni LMG 23211		ZP_14225341				
Campylobacter jejuni subsp. jejuni LMG 23223		ZP_14159503				
Campylobacter jejuni subsp. jejuni LMG 23269		ZP_14166748				
Campylobacter jejuni subsp. jejuni LMG 23263		ZP_14161069				
Campylobacter jejuni subsp. jejuni 1997-4		ZP_14194027				
Campylobacter jejuni		AAS01598				
Campylobacter jejuni subsp. jejuni 86605		ZP_14171906				
Campylobacter jejuni		BAF49092				
Campylobacter upsaliensis JV21		ZP_07893055				
Campylobacter upsaliensis RM3195		ZP_00370497				
Campylobacter lari		BAJ52736				
Campylobacter lari	clade 1	BAF48050				
Campylobacter lari	clade 1	BAH85849				
Campylobacter lari RM2100	clade 1	YP_002575942				

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Campylobacter lari	clade 1	BAF48047
Campylobacter lari	clade 1	BAJ52779
Campylobacter lari	clade 1	BAJ52796
Campylobacter coli LMG 23344		ZP_14135324
Campylobacter coli 90-3		ZP_14077429
Campylobacter coli 111-3		ZP_14076746
Campylobacter coli 1909		ZP_14111745
Campylobacter coli		BAF49128
Campylobacter coli LMG 9854		ZP_14127623
Campylobacter coli JV20		ZP_07401998
Campylobacter coli RM2228		ZP_00367553
Campylobacter coli LMG 23336		ZP_14130787
Campylobacter coli 1417		ZP_14105535
Campylobacter coli		BAJ21885
Campylobacter lari	clade 2	BAJ52762
Campylobacter lari	clade 2	BAJ52767
Campylobacter lari	clade 2	BAJ52756
Campylobacter lari	clade 2	BAJ52750
Campylobacter jejuni subsp. jejuni 1336		ZP_06373208.1
Helicobacter cinaedi ATCC BAA-847		BAM31971
Helicobacter cinaedi CCUG 18818		ZP_07806067
Helicobacter cinaedi PAGU611		YP_006235554
Helicobacter callitrichis		ADF87419
Campylobacter_fetus_subsp_fetus_82_40	clade 1	YP_891236
Campylobacter fetus	clade 1	BAF49143
Campylobacter fetus	clade 1	BAF49158
Campylobacter_fetus	clade 1	BAF33498
Campylobacter_fetus_subsp._venerealis_Azul_94	clade 1	ZP_06009829.
Helicobacter hepaticus ATCC 51449		NP_860978
Helicobacter_hepaticus		AAF19158
Helicobacter_winghamensis_ATCC_BAA_430		ZP_04583225
Helicobacter_bilis_ATCC_43879		ZP_04580438
Escherichia coli KTE11	Enterobacteriaceae clade 2	ZP_19615371
Escherichia albertii TW15818	Enterobacteriaceae clade 2	ZP_22992740
Phage cdtI Shigella dysenteriae 1012 Escherichia coli MS 21-1	Enterobacteriaceae clade 2	YP_001272541
Escherichia coli	Enterobacteriaceae clade 2	BAH78179
Escherichia coli APEC O1	Enterobacteriaceae clade 2	YP_852557
Escherichia coli STEC_C165-02	Enterobacteriaceae clade 1	ZP_12239832
Escherichia coli 53638	Enterobacteriaceae clade 1	ZP_03000678
Providencia alcalifaciens	Enterobacteriaceae clade 1	BAL72684
Providencia alcalifaciens	Enterobacteriaceae clade 1	BAL72697
Escherichia coli	Enterobacteriaceae clade 1	AAA18786.1
Escherichia coli	Enterobacteriaceae clade 1	BAH72965
Escherichia albertii TW15818	Enterobacteriaceae clade 1	ZP_22993122

[a]For pltA and pltB accession numbers are only given when these are not included in the supplemental figures.